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Evaluation of seed yield, essential oil compositions, and fatty acid profiles in advanced fennel (*Foeniculum vulgare* Mill) breeding populations

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ABSTRACT

Metabolites derived from bitter fennel serve as flavoring and healing agents in the pharmaceutical, food, and cosmetic sectors. Previously, we developed three fennel breeding populations (Early, Medium, and Late Maturity Synthetic Cultivars) with the potential to produce higher seed yield. This investigation aimed to assess the quality and quantity of the important metabolites in these three cultivars alongside their elite parental populations. Our results showed that the highest amounts of essential oil yield and seed yield were produced by, two breeding populations, Medium and Late Maturity Synthetic Cultivars. These cultivars showed positive seed yield heterosis rates of 28% and 20%, respectively, above the averages of their parental populations. GC-MS analysis of the essential oil samples identified 24 components in total. The major compounds in all genotypes were *trans*-anethole (34.09–77.84%), limonene (4.99–12.66%), methyl chavicol (1.35–38.69%), and fenchone (6–13.07%). The highest amount of *trans*-anethole belonged to Fasa and the Medium Maturity Synthetic with 77.84 % and 73.10%, respectively. The analysis of fatty acid compositions in the genotypes was conducted using GC-FID, which revealed the presence of 19 unique fatty acids, of which ten were saturated, and nine were unsaturated fatty acids. Petroselinic acid (67.11–78.44%), and linoleic acid (9.14–11.60%) were the major unsaturated fatty acids in fennel seeds. The Early Maturity Synthetic exhibited the least saturated fatty acid content (7.24%), while the highest levels of unsaturated fatty acids were found in both the Early Maturity Synthetic (89%) and Hashtgerd (88.55%). The genotypes were categorized into four groups using a heat map analysis with hierarchical clustering. Overall, the results showed that the three synthetic cultivars of fennel have outperformed the elite parental populations for seed and essential oil yields, concentration of the major industrial secondary metabolites such as *trans*-anethole, and higher proportions of unsaturated fatty acids. These cultivars show potential for usage in the pharmaceutical and healthcare industries.

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1. Introduction

Bitter fennel *(Foeniculum vulgare* Mill), a member of the Apiaceae family, is a familiar medicinal herb with a long history of usage by humans. Fennel exhibits annual, biennial, and perennial growth patterns, and its seeds, leaves, and young stems are edible (Noreen et al., 2023). Fennel seeds are rich in essential oils and fatty acids, which house valuable components utilized in the cosmetic, perfume, pharmaceutical, and food industries as flavoring and therapeutic agents (Noreen et al., 2023; Khammassi et al., 2022; Hoshang et al., 2020; Buchbauer, 2000). Fennel essential oil is primarily extracted from the seeds, with its concentration typically ranging from 2 to 4%. The major components in this essential oil are *trans*-anethole fenchone, limonene, estragole, and α-pinene (Barros et al., 2010; Shahat et al., 2011; Telci et al., 2019; Saeidniaet al., 2023). The oil content of fennel is influenced by numerous factors, including the genotypes, the specific part of the plant, its developmental stage, production method, harvest timing, and extraction techniques. Essential oil is present in varying concentrations and compositions throughout different parts of the plant (Šunić et al., 2023; Koşar et al., 2007). Several pharmaceutical benefits, including antimicrobial, antiviral, antioxidant, anti-inflammatory, anxiolytic, gastroprotective, estrogenic, cardiovascular, lipid-regulating, antidiabetic, antimutagenic, chemopreventive, anticancer, hepatoprotective, and memory-enhancing properties, have been documented in fennel. In addition to its medicinal applications, F. vulgare is utilized in various other fields such as animal husbandry (Jadid et al., 2023). Fennel seeds contain 20% oil, which makes them one of the richest plant sources of unsaturated fatty acids. Oil extracted from Apiaceae seeds usually composed of more than 50% petroselinic unsaturated fatty acid, an isomer of oleic acid. This fatty acid exhibits significant anti-inflammatory and antibacterial effects, which also prevent blood vessel blockage. It has a wide range of commercial applications in the pharmaceutical, skincare, beauty, and perfumery industries (Cosge et al., 2008; Bettaieb Rebey et al., 2016; Delbeke et al., 2016; Sayed Ahmad et al., 2018a, 2018b). Fennel oil is rich in various unsaturated fatty acids including petroselinic acid, oleic acid, linoleic acid, linolenic acid, and palmitoleic acid (Olaru and Popa, 2019; Rezaei Chiyaneh et al., 2020). Oleic acid is the most effective unsaturated fatty acid, which can lower cholesterol. Linoleic acid and linolenic acid are effective in reducing the risks of cardiovascular diseases and hypertension (Nguyen et al., 2015).

A significant portion of fennel seeds worldwide is used for extracting both volatile and nonvolatile fennel oil. The global production of fennel seeds is 842000 t/year, while Iran is the fourth largest fennel producer (FAOSTAT, 2023). For generations, Iranian farmers have been cultivating their local landraces. These landraces have evolved to thrive in specific local environments due to natural adaptations (Bogdanov et al., 2011; Ramırez Valiente et al., 2009). These landraces have very diverse morphological (Shojaiefar et al., 2015) and phytochemical properties (Salami et al., 2016; Rahimmalek et al., 2014), as well as genetic features (Maghsoudi Kelardashti et al., 2015; Shojaiefar et al., 2015).

Given the growing global demand for fennel seeds, it is crucial for breeders to pay more attention to this plant (Dashora et al., 2003). Because of the heterozygous-heterogeneous nature of the crop, self-pollination causes inbreeding depression. For crops like fennel, hybrid or synthetic cultivars are recommended as suitable breeding methods (Nematzadeh and Kiani, 2005). Compared to hybrid, synthetic cultivars offer several benefits including allowance of greater genetic diversity, less reliance on precise pollination, adaptability to various environments, and lower seed production costs (Dashora et al., 2003).

Our previous studies on Iranian fennel landraces indicated significant genetic variation (Bahmani et al., 2012, 2013), as well as agro-morphological variation (Bahmani et al., 2015, 2016). Based on their maturity habits, these fennel landraces can be classified as early, medium, or late maturity groups which need 120, 180, or 230 days from plantation to seed harvest, respectively (Bahmani et al., 2024). Also, significant diversity in terms of fatty acid composition among these fennel landraces has been reported (Izadi-Darbandi et al., 2023). In a different study, a wide range of drought tolerance among these landraces has been identified (Baghcheghi, 2012). This diversity among these landraces provides a valuable opportunity for breeders to develop high-yielding and stress-tolerant cultivars.

Recently, our research team developed fennel breeding populations, comprising three high-yielding synthetic cultivars obtained by crossing genotypes with a high general combining ability for seed yield and essential oil content (Bahmani et al., 2023). Preliminary investigations revealed that these synthetic cultivars outperformed their parents in seed yield in drought conditions while showing similar performance to the parents under normal growth conditions (Akbari et al., 2015a). Further studies showed that these three synthetic cultivars have higher antioxidant activity, flavonoid content, phenolic content, and phenolic compounds (such as chlorogenic acid and ascorbic acid) (Akbari et al., 2023a), indicating that synthetic cultivars of fennel can be used as a natural antioxidant source in industry.

Here in this study, our primary goal was to thoroughly examine critical characteristics such as seed yield, quality and quantity of essential oils compositions, and fatty acid profiles of the second generation (Syn^2) of the three fennel synthetic cultivars. Furthermore, a comparison was made between these cultivars and their elite parent populations. Our findings offer valuable insights into the performance of synthetic cultivars, particularly in crops such as fennel.

2. Materials and methods

This study was conducted at the agriculture investigation facility of the College of Aburaihan, University of Tehran, located in Pakdasht, Iran. The farm is positioned at the coordinates of 35˚29′ N latitude and 51˚40′ E longitude: 35˚29′ N and 51˚40′ E, with an estimated elevation of 1026 m above sea level. The region has a semi-arid climate with an average of 175 mm per year of precipitation and temperatures averaging 16.9 ˚C.

2.1. Plants material

Three synthetic cultivars of fennel (*Foeniculum vulgare* Mill.) were developed by the fennel breeding program of the University of Tehran. The three synthetic cultivars were Early Maturity Synthetic Cultivar, Medium Maturity Synthetic Cultivar, and Late Maturity Synthetic Cultivar, and they were generated by crossing 10, 10, and 5 parental populations respectively with high general combining ability for seed yield and essential oil content. This study assessed these breeding populations, along with the selected elite parental populations: Early Maturity Synthetic with two parental populations (Hashtgerd and Fasa), Medium Maturity Synthetic with four parental populations (Moghan, Fozve, Meshkinshahr, and Khash), and Late Maturity Synthetic with two parental populations (Chahestan and Hajiabad) (Table 1).

2.2. Field experiment

For the open field assessment, a randomized complete block design (RCBD) with three repetitions was used. Within each replication, genotypes including synthetic cultivars and elite parental populations, were planted in plots measuring 1.5 by 1.5 m. The row spacing was set to 30 cm, and the seeds were planted at a depth of 2–3 cm. This study was conducted in two successive years 2018 and 2019, after seed plantation in March of each year. The plant density was regulated by thinning the plants to achieve a final density of 10 plants per square meter (Khorshidi et al., 2010). We assessed traits related to yield and its components, such as number of branches (NB), number of nodes (NN), number of umbels (NU), number of umbellets per umbel (Numb), number of seeds per umbel (NSU), and number of seeds per umbellet (NSumb) directly in the field. Following this, the seeds from each plot were harvested separately to measure seed yields, calculated as the total weight of seeds per square meter. Essential oil compositions and fatty acid profiles were analyzed on samples collected during the second year of the field trial (2019).

2.3. Extraction of essential oil

Essential oils were extracted from fully ripe seeds, and for this purpose, 20 gr of seed from each genotype was milled to a fine powder and then mixed with 500 mL of deionized water and heated to 100 °C. Subsequently, the oil was collected using a Clevenger-type apparatus for 2 h (Akbari et al., 2015b, 2023b). There were three replications of each extraction. Later on, essential oil yield was calculated as follows: (Seed yield × essential oil content)/100 (Ramadan, 2007).

2.4. GC–MS analysis of essential oils

Essential oils were analyzed using gas chromatography combined with mass spectrometry (GC/MS) with a Varian CP-3800 instrument and a VF-5 column (30 m in length, 0.25 mm inner diameter, 0.25-μm film thickness). The carrier gas was helium, flowing at a rate of 1 ml/min. The ionization energy used by the mass spectrometer was 70 electron volts. The injector and detector temperatures were kept at 250 °C, and the oven temperature was programmed to ramp up from 60 to 250 °C at a rate of 3 °C per minute. The injection volume was 1 μL in the split mode. By calculating the retention indices of the essential oil at certain temperature settings and using n-alkanes with a range of C6 to C24 as benchmarks, the composition of the oil was evaluated. Utilizing a VF-5 column and the same chromatographic parameters, the oils were examined. The chemicals were identified by comparing their mass spectra to those in the internal reference mass spectra library (Wiley 7) or those of authentic compounds. Additionally, the retention indices of the chemicals were compared to either recognized compounds or values listed in the published literature to confirm their identity. Relative area percentages discovered via Flame Ionization Detection (FID) were used for quantification without the need for correction factors.

2.5. Total lipid extraction

Approximately 10 g of ground seeds underwent extraction in 100 mL of petroleum ether for 2 h employing an automated Soxhlet device (Gerhardt Soxtherm). The remaining material was desiccated until a stable weight was attained. Potassium hydroxide (KOH) in methanol was used for the methylation process instead of BF_3 before the analysis (AOAC, 1990).

2.6. Analysis of fatty acid compounds by GC-FID

The fatty acids examination were done by gas chromatography (Clarus 500), an autosampler (PerkinElmer, Shelton, CT, USA), and a detector for flame ionization. A fused-silica capillary SGE column (100 m × 0.32 mm, ID 0.25 μm, BP20 0.25 UM; PerkinElmer, Austin, TX, USA) was used for this analysis. The oven was initially reheated for 5 min at 140 C. The temperature was

Table 1
Fennel genotypes and synthetic cultivars used in the experiment.

Synthetic cultivars	Parents of synthetic cultivars	Genotype	Altitude (m)	Latitude	Longitude
Early Maturity Synthetic (Early Syn)	Parents of Early Maturity Synthetic	Fasa	1288	28°58′N	53°41′E
		Hashtgerd	1426	35°65′N	50°43′E
Medium Maturity Synthetic (Medium Syn)	Parents of Medium Maturity Synthetic	Moghan	31	39°39′N	47°55′E
		Meshkinshahr	1568	38°23′N	47°40′E
		Fozveh	1612	32°36′N	51°26′E
		khash	1394	28°13′N	61°12′E
Late Maturity Synthetic (Late Syn)	Parents of Late Maturity Synthetic	Hajiabad	931	28°19′N	55°55′E
		Chahestan	27	27°13′N	56°22′E

then increased to 200 °C by adding 4 °C per minute, and then it was raised to 220 °C by adding 1 °C each minute. Temperature adjustments were made to the injector and detector for 220 °C and 280 °C, respectively. The carrier gas pressure was maintained at 16 psi, and the sample volume was limited to 1 μL. It was a 1:100 split. To identify the fatty acids, a mixture of 37 fatty acid methyl esters (FAMEs) from Supelco in Bellefonte, Pennsylvania, was used (Kafkas et al., 2017).

2.7. Statistical analyses

The analysis of variance was performed utilizing SAS statistical software, and the genotypic means were compared using the Duncan test at a significance level of $P < 0.05$. Using R Studio (version 4.2.3), we conducted principal component analysis, cluster heat map analysis, and Pearson's correlations.

3. Results

3.1. Yield and yield component

To evaluate the effects of year, genotype, and their interactions on the seed yield and yield components in fennel genotypes, a combined variance analysis was conducted (Table 2). The results showed significant differences in seed yield and yield components between genotypes and years. According to the experiment results, characteristics like the number of nodes, number of seeds per umbel, and essential oil percent were not significantly influenced by genotype × year interactions. However, substantial differences were seen at a 1% significance level for other characteristics. Based on the mean comparisons (Table 3), it is evident that in the first year, the Medium Maturity Synthetic with Moghan and Fozveh genotypes exhibited notably higher seed yields, measuring 235, 229, and 213 g/m^2, respectively. In the second year, the Medium Maturity Synthetic, Late Maturity Synthetic, and Fozveh genotypes demonstrated the highest seed yields, registering 266, 260, and 217 g/m^2, respectively (Fig. 1). For the cumulative seed yield of both years, the Medium Maturity Synthetic and Fozveh genotypes stood out with the highest seed yields, recording 251.19 and 215.55 g/m^2, respectively, while the Chahestan genotype displayed the lowest seed yield at 144.06 g/m^2 (Table 3). The calculated heterosis rates between the synthetic cultivars and their parental populations, determined by averaging the parental values, indicated a heterosis range of 19.86%–28.21% for the seed yield trait (Table 4). Notably, the Late Maturity Synthetic genotype exhibited the lowest heterosis, while the Medium Maturity Synthetic demonstrated the highest rate.

Among the genotypes, Hashtgerd and Late Maturity Synthetic exhibited higher essential oil content at 2.2 and 2.08 ml, respectively, in the first year. Fozveh, Moghan, and Early Maturity Synthetic showed higher essential oil content at 2.08, 2.04, and 2.01 ml dried matter, respectively in the second year. When considering the two-year average for essential oil content, the fennel genotypes with the highest values were Hashtgerd, Fozveh, and Moghan, recording 2.07, 2.02, and 2.0 ml, respectively. The heterosis rates for this trait indicate that the Late Maturity Synthetic displayed the highest level of heterosis, reflecting a notable 7.94% increase (Table 4).

In terms of essential oil yield, the Medium Maturity Synthetic and Fozveh showcased the highest levels of essential oil yield at 4.69 and 4.19 ml/m^2, respectively in the first year. Meanwhile, in the second year, the top performers among fennel genotypes were the Medium Maturity Synthetic and Late Maturity Synthetic, recording 5.02 and 4.60 ml/m^2, respectively. Notably, the Medium Maturity Synthetic emerged as the best fennel genotype, averaging a yield of 4.83 ml/m^2 when considering the combined essential oil yield over the two years. The heterosis rate for the essential oil yield trait across the three synthetic genotypes was 27.15%. Notably, the Late Maturity Synthetic genotype displayed the highest heterosis rate (29.21%) (Table 4).

3.2. Correlations between yield and yield component

According to the average values from the evaluations performed out in 2018 and 2019 fennel genotypes, relationships between seed yield and yield component were investigated (Fig. 2). Numerous variables, such as the number of seeds per umbel (r = 0.92), number of umbels (r = 0.75), number of umbellets (0.77), number of seeds per umbellet (r = 0.67), and yield of essential oils (r = 0.91), showed strong positive associations with seed yield.

Table 2
Two-year pooled analysis of variance (ANOVA) for seed yield and related components in fennel genotypes.

SOV	DF	No. umbel	No. seed per umbel	No. umbellets per umbel	No. seed per umbellet	No. branches	No. nodes	Essential oil content (cc)	Essential oil yield (ml/m^2)	Seed yield (gr/m^2)
Year	1	57712.36**	31614.85**	0.89 ^{ns}	2230.69 **	100.27**	6.58**	0.069 ^{ns}	5.70**	10458.26**
Rep	4	23.66 ^{ns}	478.45 ^{ns}	0.41 ^{ns}	2.70 ^{ns}	0.27 ^{ns}	1.22 *	0.063 *	0.17 ^{ns}	337.38 ^{ns}
Genotype	10	689.86 **	60462.83**	99.48**	46.50 **	14.75 **	14.17 **	0.093 **	4.43 **	11223.20**
Year* Genotype	10	418.30**	3837.87**	11.44**	20.19 ^{ns}	2.78 **	0.87 ^{ns}	0.029 ^{ns}	0.88 **	2317.24**
Error	40	53.32	1189.85	2.63	13.76	0.46	0.42	0.019	0.13	199.32
Mean	–	58.06	371.79	21.11	25.97	9.61	10.16	1.93	3.35	172.9
CV (%)	–	12.57	9.27	7.68	14.28	7.073	6.44	7.21	11.05	8.16

*, **Significant at 0.05 and 0.01 P levels, respectively and ns = non-significant.

Table 3
Comparison of mean seed yield and yield components in fennel genotypes over two years.

Genotype	No. seed per umbellet		No. umbellets per umbel		No. umbel		No. seed per umbel		No. branches		No. nodes		Essential oil content (cc)		Essential oil yield (ml/m^2)		Seed yield (gr/m^2)	
	Y1	Y2	Y1	Y2	Y1	Y2	Y1	Y2	Y1	Y2	Y1	Y2	Y1	Y2	Y1	Y2	Y1	Y2
Fasa	18.00b	30.22 ab	19.33de	19.22b	20.78f	66.67c	256.00c	316.00e	6.00f	9.00c	7.33d	7.78e	1.71cd	1.95abc	1.89d	2.69c	110.10c	134.62e
Moghan	21.66a	31.89ab	23.89ab	26.67a	35.67b	88.22b	500.33a	446.83bc	8.44c	10.56b	10.89b	9.44 d	1.96abc	2.04abc	4.53a	3.65 b	229.73a	179.01d
Meshkinshahr	22.22a	29.22ab	23.45abc	21.33b	34.00bc	69.33bc	365.33b	421.17bc	7.78cde	11.44ab	10.89b	10.67bc	1.63d	1.70c	2.98bc	2.88c	182.00b	169.98d
Fozveh	23.33a	30.00ab	23.89ab	26.67a	32.00bc	80.22bc	386.33b	458.67ab	8.00cd	11.78a	11.56ab	10.22cd	1.96abc	2.08a	4.19a	4.52a	213.33a	217.21bc
Hajiabad	18.00b	34.00ab	17.33ef	13.78c	29.33de	106.78a	381.33b	441.33bc	10.00b	11.67a	12.33a	12.33a	1.70cd	1.70c	2.68bcd	3.98 b	157.63b	209.48c
Hashtgerd	18.33b	30.67ab	19.00de	19.11b	22.11f	77.00gc	233.33c	353.00ed	6.45ef	8.44c	8.67c	7.61e	2.20a	1.95abc	1.67d	2.69c	107.03c	127.83f
Early Maturity Synthetic	22.11a	35.22ab	20.44cde	18.67b	21.56f	77.00bc	292.67c	396.00cd	6.78def	9.22c	8.00cd	8.33e	1.86bcd	2.01abc	2.20cd	3.64b	121.31c	161.58d
Medium Maturity Synthetic	23.67a	36.00a	25.56a	27.33a	38.67a	115.67a	518.33a	510.08a	7.33cde	11.56ab	11.33ab	11.44ab	1.98abc	1.76bc	4.69a	5.02a	235.70a	266.68a
Late Maturity Synthetic	16.44b	34.45ab	20.44cde	21.33b	27.33e	117.00a	384.33b	436.33bc	11.67a	11.89a	11.56ab	10.56bcd	2.08ab	1.80bc	2.63cd	4.60a	126.22c	260.59 b
Khash	25.00a	32.00ab	21.89abc	27.00a	34.33bc	82.89bc	380.67b	401.75bcd	7.78cde	11.44ab	11.00ab	9.67cd	1.95abc	1.90abc	3.43b	3.93b	175.73b	199.53c
Chahestan	13.00c	26.00b	15.78f	12.44c	17.66g	87.22b	150.00d	149.33f	12.00a	12.33a	11.78a	10.33bcd	1.90bcd	2.0 ab	1.99d	2.53c	105.0c	123.11e

In each character, genotypes with one common letter at least are not significantly difference at the 5% level.

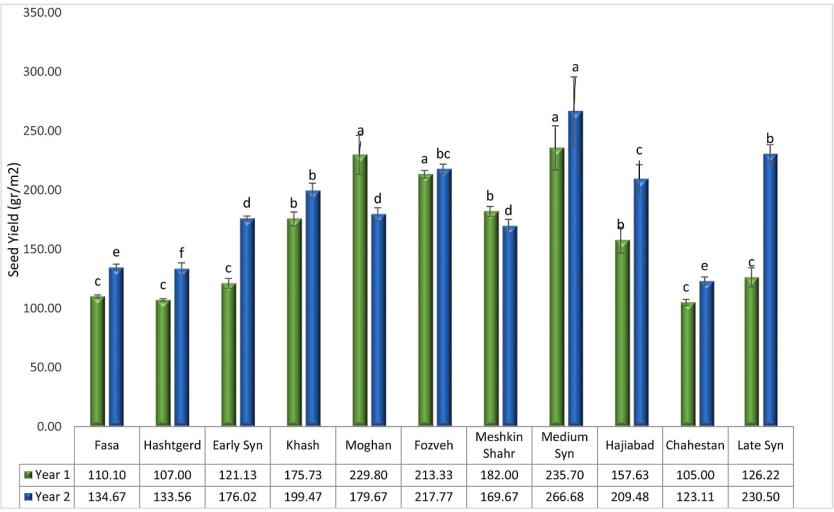


Fig. 1. Comparative seed yield of fennel genotypes over two years.

Table 4
Mean and percentage of heterosis in seed yield and yield components among fennel genotypes based on two-year data.

	No. branches	No. seed per umbel	No. umbel	No. nodes	No. umbellets per umbel	No. seed per umbellet	Essential oil content (cc)	Essential oil yield (ml/m^2)	Seed yield (gr/m^2)
Early Maturity Synthetic	8.00	344.33	47.28	8.17	19.56	28.67	1.97	2.95	148.57
Parents Mean of	7.47	289.58	46.64	7.85	19.17	24.31	1.98	2.40	121.33
Heterosis	7.06	18.91	1.37	4.07	2.03	17.94	−0.42	23.06	22.45
Medium Maturity Synthetic	9.44	514.21	77.17	11.39	26.44	29.83	1.93	4.86	251.19
Parents Mean of	9.65	420.18	57.08	10.54	24.35	26.92	1.91	3.76	195.93
Heterosis	−2.16	22.38	35.18	8.04	8.61	10.84	1.05	29.02	28.21
Late Maturity Synthetic	11.78	410.33	72.17	11.06	20.89	25.44	2.04	3.62	178.36
Parents mean of	11.50	280.50	60.25	11.69	14.83	22.75	1.89	2.80	148.80
Heterosis	2.42	46.29	19.78	−5.46	40.82	11.84	7.94	29.21	19.86

3.3. Cluster analysis for yield and yield component

To classify genotypes according to yield and yield component, cluster analysis was performed; the outcomes are shown in Fig. 3. Three separate clusters were formed from the genotypes. The first cluster included the four genotypes of the Medium Maturity Synthetic and three of its studied parental populations (Moghan, Fozveh, and Khash). Compared to the other two, this cluster had the highest seed yield and yield components. The second cluster included the Late Maturity Synthetic and its two studied parental populations (Hajiabad and Chahestan), plus Meshkinshahr (a medium maturity parent), and comparatively had moderate values for the majority of the studied traits, they notably showed the highest numbers for both the number of nodes and branches. The third cluster included Early Maturity Synthetic and its two studied parental populations (Fasa and Hashtgerd) and comparatively had the lowest values for the yield and yield components.

3.4. Essential oil composition

Utilizing gas chromatography coupled with mass spectrometry (GC/MS), in total we identified 24 components in fennel essential oil, collectively constituting 97.92% of the total essential oil. As depicted in Table 5, the fennel genotypes exhibit a range of *trans*-anethole concentrations from 34.32% to 78.19%, methyl chavicol from 5.59% to 38.69%, fenchone from 6% to 13.7%, and limonene from 4.99% to 12.66% (Table 5). This wide variability in fennel compositions is attributed to genetic differences. The highest percentage of *trans*-anethole belonged to Fasa, and the Medium Maturity Synthetic (77.84 and 73.10%, respectively). The lowest percentage was obtained in Hajiabad (34.09%). Hajiabad and Khash genotypes had the highest concentration of limonene (12.66 and 10.35%). Among the three synthetic cultivars, the Early Maturity Synthetic cultivar had the highest amount of limonene (9.62%). Hajiabad had the highest amount of methyl chavicol (38.69%) compared to other genotypes, and Chahestan had the lowest amount (1.35%) of this compound (Table 5). The percentage of fenchone in Khash (13.07%) was higher than in other genotypes, while Fasa had the lowest (6%).

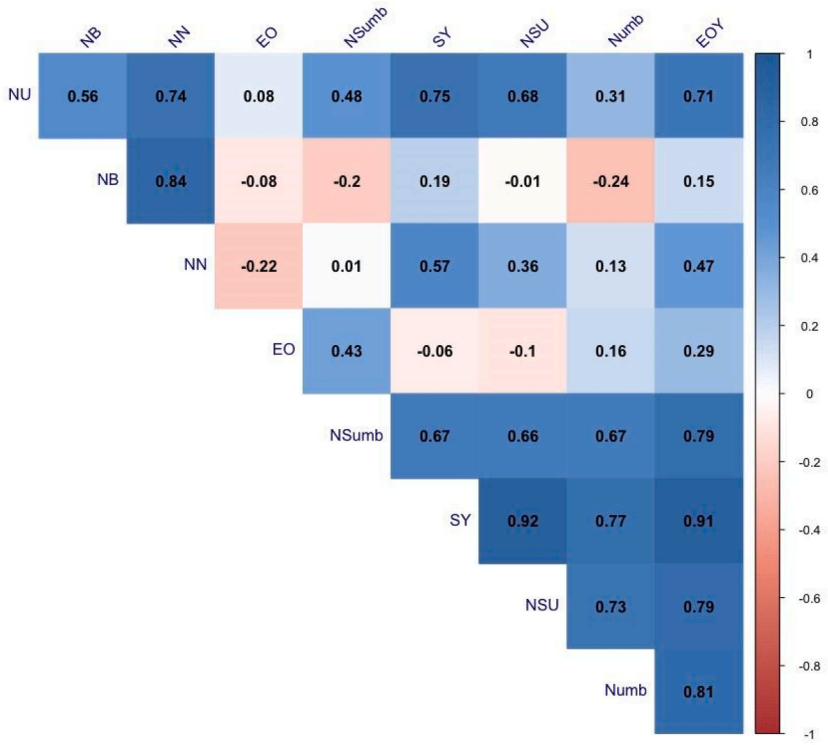


Fig. 2. Phenotypic correlation coefficients among seed yield and yield components in fennel genotypes over two years.

3.5. Fatty acids composition

Based on the results of the fatty acid assessment by gas chromatography (GC-FID), 19 fatty acids were identified in fennel seeds (Table 6, Fig. 4). The most important known fatty acids are petroselinic acid (C18: 1 (n-12)) and linoleic acid (C18:2n6c). The amount of saturated fatty acids (SFA) varied between 7.24 and 22.63%. The lowest saturated fatty acid was observed in Early Maturity Synthetic. The main saturated fatty acid among the studied genotypes was lauric acid (C12: 0), which varied between 0.19 (Moghan) to 15.07 (Khash). The fennel oil also included margaric acid (C17: 0), palmitic acid (C16: 0), stearic acid (C18: 0), myristic acid (C14: 0), and arachidonic acid (C20: 0), which was observed in all genotypes but with different amounts (Table 6). Unsaturated fatty acids (UFA) amounts varied between 69.87 (Khash) to 89.00% (Early Maturity Synthetic). Petroselinic acid percentage ranged from 60.11% (Khash) to 78.44% (Early Maturity Synthetic). The highest amount of oleic acid was present in Late Maturity Synthetic (0.37%). The amount of linoleic acid (omega 6), as a fatty acid with two unsaturated bands, ranged from 8.70 to 11.60 %, with the Late Maturity Synthetic having the highest and Khash having the lowest amount. In the case of polyunsaturated fatty acids, the amount of γ-linolenic acid (omega 3) in the fennel genotypes ranged from 0.12 to 0.42, and the Late Maturity Synthetic showed the highest amount (Table 6).

The ratio between polyunsaturated fatty acids and saturated fatty acids (PUFA/SFA) in vegetable oils is considered an important parameter in determining the nutritional value of oil. If this index is higher than 1, the oil has a high nutritional value. The ratio of polyunsaturated to saturated fatty acids among the eleven genotypes varied from 0.43 to 1.59, and the highest ratios belonged to the Late Maturity Synthetic, Fozveh, and Early Maturity Synthetic. Since most genotypes showed a rate higher than 1 for this index, fennel oil can be considered an oil with high nutritional value.

3.6. PCA analysis

Principal Component Analysis (PCA) was employed for the essential oil content, main essential oil composition, and essential oil yield to categorize and distinguish fennel genotypes. The findings showed that the first two components accounted for 68.2% of the overall variation. The first PC (PC1) had positive associations with α-pinene, limonene, methyl chavicol, β-myrcene, essential oil, and essential oil yield, and it explained 39.6% of the overall variation. Despite this, it correlated negatively with *trans*-anethole and α-phellandrene. 28.6% of the overall variance was explained by PC2. The biplot based on these traits demonstrated that the genotypes were divided into three groups (Fig. 5). Group 1 had just one genotype (Hajiabad), which was placed on the positive side of PC1, methyl chavicol was discovered to be the prominent constituent in this genotype essential oil. Group 2 included Fozveh, Hashtgerd, Early Maturity Synthetic, Late Maturity Synthetic, and Khash, which were identified by higher limonene and essential oils yield, as well as greater amounts of fenchon and essential oil content. The four genotypes that formed Group 3 were dispersed on the PC1 negative side. Moghan, Meshkinshahr, Chahestan, Medium Maturity Synthetic, and Fasa are composed of a single group with high *trans*-anethole and low methyl chavicol.

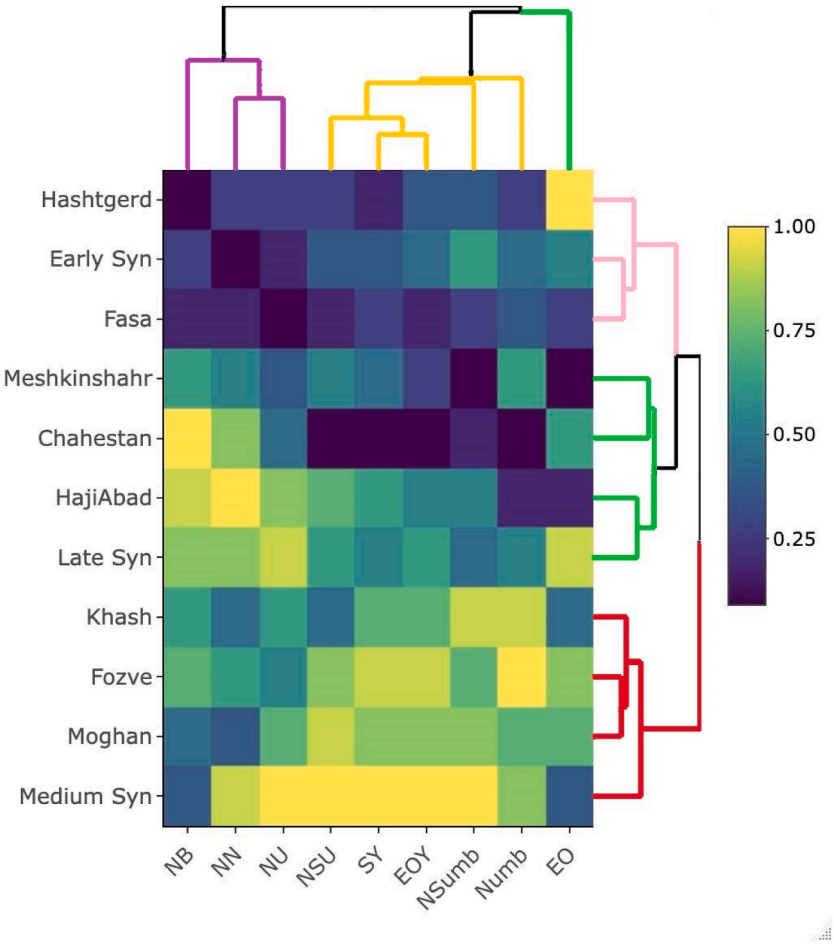


Fig. 3. Heat map analysis (HMA) of seed yield and yield components, of fennel genotypes.

A PCA on fatty acid components was also carried out to detect relationships between genotypes and fatty acid profiles. According to the findings, the first and second components accounted for 69.7% of the overall variation. Petroselinic acid, palmitic acid, linoleic acid, linolenic acid, stearic acid, and oleic acid, all had positive correlations with PC1, which exhibited 47.1% of the overall variation. In the PC2 findings, lauric acid, and myristic acid were highly correlated. The biplot of PC1 and PC2 was created to categorize the genotypes using the PCs (Fig. 6). The biplot separates fennel genotypes into three groups. Moghan, Fozve, Meshkinshahr, Hashtgerd, Early Maturity Synthetic, Late Maturity Synthetic, and Hajiabad are members of group 1, which were identified by a high level of petroselinic acid, oleic acid, linoleic acid, and palmitoleic acids. Group 2 included genotypes Medium Maturity Synthetic and Khash, characterized by elevated levels of lauric acid and myristic acid. In Group 3, only the genotype Chahestan was found, positioned favorably on PC2. Notably, pentadecenoic acid and *cis*-10-pentadecanoic acid were identified as significant components in the oil of this genotype.

3.7. Heat map analysis

Hierarchical cluster heat map analysis is a technique to visually represent a group of samples and the corresponding measured substances. In this study, hierarchical cluster heat map analysis was conducted using the primary constituents of essential oils, the overall essential oil content, yield, and the fatty acid profile of fennel genotypes. After conducting the analysis, they were categorized into four clusters (Fig. 7). The first cluster which was composed of only the Khash genotype was characterized by the highest levels of α-pinene, fenchone, β-myrcene, lauric acid, and pentadecenoic acid, while it exhibits the lowest amounts of petroselinic acid, linoleic acid, and linolenic acid. The second cluster, composed of Hajiabad and Late Maturity Synthetic, was characterized by elevated levels of methyl chavicol, limonene, *cis*-10-pentadecanoic acid, oleic acid, and linoleic acid. The third cluster consisted of the Chahestan genotype exhibited the highest levels of stearic acid and α-phellandrene and the lowest level of oleic acid. From a health perspective, the most noteworthy cluster included Fozveh, Mogham, Hashtgerd, Fasa, Early Maturity Synthetic, and Medium Maturity Synthetic. These genotypes were distinguished by their high levels of petroselinic acid, linoleic acid, linolenic acid, *trans*-anethole, essential oil yield, and low levels of α-pinene, fenchon, β-myrcene, lauric acid, and pentadecenoic acid. Given the advantageous properties of pet-

Table 5
Analysis of essential oil composition in fennel genotypes for the second year.

Genotype	RI	Fasa	Moghan	Meshkin shahr	Hajiabad	Hashtgerd	Fozveh	Chahestan	Early Maturity Synthetic	Medium Maturity Synthetic	Late Maturity Synthetic	Khash
Monoterpene hydrocarbons												
α-Pinene	936	0.57	0.68	0.58	0.55	1	1.25	0.89	0.95	0.82	1.26	1.23
Camphene	954	0.07	0.08	0.07	0.1	0.15	0.11	0.08	0.12	0.09	0.22	0.25
Sabinene	976	0.13	0.19	0.18	0.23	0.19	0.25	0.23	0.24	0.22	0.33	0.26
β -Pinene	983	–	0.05	–	0.04	0.05	0.13	0.03	0.08	0.06	0.07	0.09
β -Myrcene	991	0.31	0.41	0.4	0.57	0.61	0.52	0.31	0.58	0.42	0.75	0.96
α-Phellandrene	1011	0.08	0.1	0.09	0.1	0.14	0.09	0.41	0.15	0.11	0.16	0.18
Ortho -Cymene	1028	0.06	0.31	0.6	1.28	0.11	1.52	0.17	0.22	0.41	1.26	0.53
Limonene	**1030**	**4.99**	**8.2**	**7.64**	**12.66**	**8.46**	**8.36**	**5.9**	**9.62**	**7.38**	**8.78**	**10.35**
β-Ocimene	1032	0.62	0.94	0.79	0.65	1.07	0.78	1.3	1.19	0.84	0.93	1.06
Linalool	1045	–	–	–	–	–	–	–	–	–	0.04	–
α-Ocimene	1054	–	–	–	–	0.05	–	–	0.05	–	–	0.05
gama-Terpinene	1062	0.35	0.73	0.73	1.95	0.39	0.78	0.85	0.38	0.76	1.36	0.99
Oxygenated monoterpenes												
Fenchon	**1096**	**6**	**7.77**	**7.26**	**8.14**	**9.34**	**9.26**	**8.2**	**8.9**	**7.11**	**11.02**	**13.07**
allo-ocimene	1128		0.06			0.06			0.07	0.05	0.05	0.06
Camphor	1154	0.22	0.24	0.28	0.25		0.34	0.28	0.29	0.28	0.4	0.52
β-Terpinolen	1144	–	–	–	–	0.29		0.03	–	–	0.05	–
Methyl chavicol	**1205**	**7.36**	**6.61**	**7.09**	**38.69**	**7.76**	**6.72**	**15.06**	**7.52**	**7.13**	**18.86**	**5.59**
Fenchyl acetate	1222	–	–	0.32	0.17	–		0.37	–	–	–	–
p-Anis aldehyde	1253		0.22	0.75	–	–	0.2		–	0.36	–	–
Iso-bornyl acetate	1287	0.29	0.14	0.05		0.21	0.15	0.4	0.12	0.12	0.07	0.23
Trans-Anethole	**1299**	**78.19**	**72.87**	**72.8**	**34.32**	**69.73**	**69.32**	**65.43**	**69.33**	**73.44**	**54.09**	**63.5**
α -Cubebene	1345	0.7	0.4	0.27	0.23	0.39	0.15	0.03	0.04	0.4	0.23	0.34
Sesquiterpene hydrocarbons												
Carvotanacetone	1347	–	–	0.1	0.07	–	0.07	–	0.08	–	–	–
α-Copaene	1381	0.06	–	–	–	–	–	0.03	0.07	–	0.07	–

roselinic acid, linolenic, and linoleic acids, as well as *trans*-anethole, it is advisable to explore synthetic cultivars as excellent sources for trait discovery on health and flavor-related traits.

4. Discussion

This study conducted a comprehensive assessment of fennel breeding populations. The objectives were to evaluate the second generations of three synthetic cultivars compared to some of their elite parental populations and characterize their agronomic, phytochemical, and nutritional traits.

4.1. Yield and yield components

The analysis of variance across two years of field trials revealed a profound and statistically significant impact of both year and genotype on all observed traits. The interaction between these factors significantly influenced most of the traits studied. Lal (2008) and Shojaiefar et al. (2015) previously observed substantial variability among fennel genotypes and across years, especially for economically important characteristics including yield.

Meena et al. (2010) reported considerable diversity for yield and its components in fennel ecotypes. Similarly, Safaei et al. (2013) identified significant trait variations among 12 fennel ecotypes, and Kumar et al. (2017) noted disparities in morphological features and yield characteristics within the fennel germplasm.

In our study here, the synthetic cultivars across the two years exhibited average seed yields ranging from 141.44 to 251.19 g/m^2, surpassing the average values for parental populations which ranged from 117.43 to 215.27 g/m^2. The Medium Maturity Synthetic cultivar particularly excelled with the highest yield of 251.19 g/m^2. Positive heterosis effects were evident across most characters, showcasing increased vigor in the synthetic cultivars. The substantial 20–28% improvement in yield over parental means further emphasized the success of the synthetic cultivars. Seed yield records in this study were relatively similar to those in Iranian fennel landraces which ranged from 146 to 254 g/m^2 (Bahmani et al., 2015), and outperformed several Indian cultivars whose yield ranged from 112 to 178 g/m^2 (Kumar et al., 2017); this underscores the promising potential of synthetic cultivars as an efficient breeding approach for fennel.

The essential oil yields across our fennel genotypes ranged from 2.14 to 4.85 mL/m^2 over two years. The Medium Maturity Synthetic cultivar notably outperformed, achieving the maximum yield of 4.85 mL/m^2, the synthetic cultivars indicate a remarkable 23–29% increase over parental means. This suggests strategic crossing led to the attainment of significant heterosis.

Table 6
Analysis of fatty acid compositions in fennel genotypes for the second year.

Composition		RT	Fasa	Hashtgerd	Early Maturity Synthetic	Khash	Meshkin shahr	Fozveh	Moghan	Medium Maturity Synthetic	Chahestan	Hajiabad	Late Maturity Synthetic
Caprylic Acid	C8:0	5.731	0.00	0.02	0.01	1.61	0.01	0.02		0.50	0.02	0.04	0.02
Capric Acid	C10:0	7.003	0.02	0.02	0.03	0.08	0.03	0.04	0.02	0.29	0.01	0.26	0.04
Lauric Acid	C12:0	8.223	1.73	2.24	0.25	15.07	2.86	0.63	0.19	9.81	1.23	3.79	0.32
Myristic Acid	C14:0	9.734	0.23	0.20	0.12	1.09	0.21	0.07	0.12	0.69	0.06	0.76	0.18
Pentadecenoic Acid	C15:0	10.460	0.05	0.02	0.50	0.08	0.02	0.06	0.08	0.08	0.15	0.63	0.03
Palmitic Acid	C16:0	11.501	3.82	4.41	3.82	2.95	4.00	4.08	4.39	3.54	3.76	3.71	4.38
Margaric Acid	C17:0	12.700	0.26	0.06	0.03	0.15	0.04	0.19	0.31	0.03	0.05	0.09	
Stearic Acid	C18:0	14.141	1.23	1.32	1.25	0.99	1.20	1.22	1.32	1.06	2.22	1.26	1.46
Arachidic Acid	C20:0	17.274	0.28	0.28	0.28	0.18	0.26	0.25	0.26	0.21	0.11	0.20	0.27
Behenic Acid	C22:0	22.183	0.71	0.76	0.95	0.42	0.48	0.85	1.09	0.40	0.34	1.24	0.86
Saturated fatty acids (SFA)			8.33	9.33	7.24	22.63	9.12	7.39	7.76	16.60	7.96	11.99	7.54
Myristoleic Acid	C14:1	9.991	0.05	0.06	.	0.08	.	0.04	0.03	0.69	0.04	0.06	0.06
cis-10-pentadecanoic acid	C15:1	10.814	0.19	0.13	0.07	0.16	0.10	0.09	0.15	0.11	0.06	0.83	0.19
Palmitoleic Acid	C16:1	12.035	0.06	0.05	0.04	0.05	0.15	0.07	.	0.05	0.05	0.11	0.05
Oleic acid	(C18:1 n9c)	14.442	0.32	0.35	0.32	0.21	0.32	0.35	0.10	0.28	0.10	0.30	0.37
Petroselinic acid	**(C18:1 n12)**	**14.761**	**75.50**	**76.78**	**78.44**	**60.11**	**74.86**	**73.20**	**74.09**	**71.33**	**74.77**	**69.51**	**72.97**
Linoleic acid	**(C18:2n6c)**	**15.59**	**9.88**	**10.34**	**9.61**	**8.70**	**10.56**	**10.30**	**10.21**	**8.85**	**9.14**	**9.45**	**11.60**
γ-linolenic acid	C18:3	16.36	0.24	0.25	0.22	0.12	0.23	0.24	0.25	0.19	0.18	0.21	0.42
Gonodic Acid	C20:1	17.73	0.28	0.11	0.05	0.17	0.05	0.13	0.07	0.03	0.06	0.24	0.05
Eruric Acid	C22:1	23.32	0.29	0.48	0.25	0.27	0.30	0.23	0.37	0.21	0.32	0.35	0.41
Unsaturated fatty acids (UFA)			86.82	88.55	89.00	69.87	86.59	84.65	85.26	79.74	84.72	81.04	86.10
Sum			95.15	97.88	96.24	93.50	95.71	92.04	93.40	96.64	92.69	93.03	93.65
MUSA			76.70	77.96	79.17	61.06	75.79	74.11	74.80	69.70	75.40	71.38	74.08
UFA/SFA			10.43	9.49	12.28	3.13	9.50	11.45	10.48	4.72	10.64	6.76	11.42
PUSA			10.12	10.59	9.82	9.82	10.79	10.54	10.46	10.04	9.32	9.66	12.02
PUFA/SFA			1.22	1.14	1.36	0.43	1.18	1.43	1.29	0.59	1.17	0.81	1.59

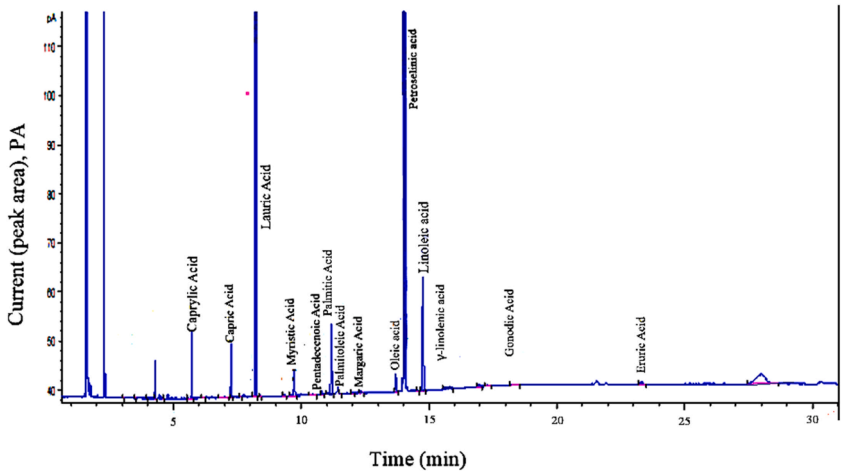


Fig. 4. Chromatogram GC-FID of seeds fatty acids of fennel.

Essential oil content recorded here from our study in two years ranged from 1.70 to 2.08% of dry matter. This variability aligns with findings from previous studies, where essential oil contents were reported to be 1.86–6.4% (Safaei et al., 2013), 2.27%–5.94% (Bernath et al., 1996) and 1.2-5.06% (Khammassi et al., 2018).

Assessing the genotypes across multiple years was a great way to increase the reliability of the data. This way of evaluation facilitated the examination of the unpredictable aspects of genotype-environment interactions, primarily influenced by annual effects (Lin and Binns, 1988). In this study, the genotype-by-year interaction (G × Y) exhibited high significance across most of the studied traits,

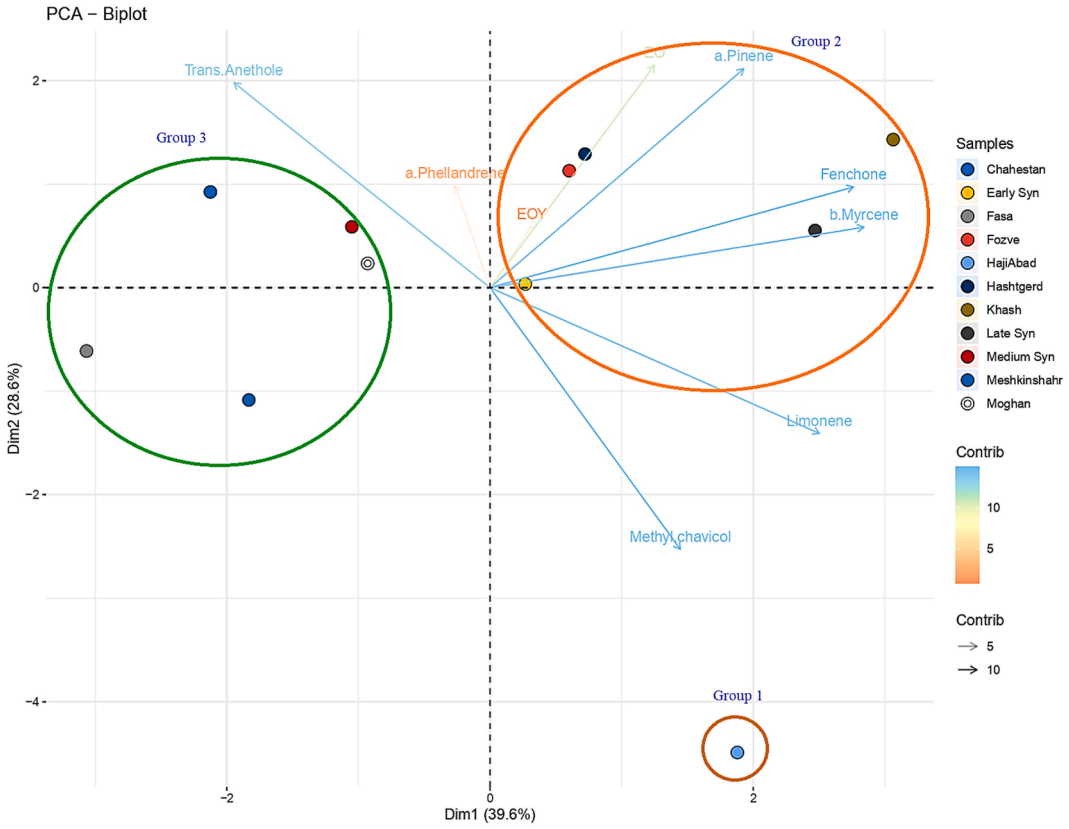


Fig. 5. Principle component analysis for main essential oil components, essential oil, and essential oil yield of fennel genotypes.

signifying considerable variations in the performance and ranking of genotypes in different years, specifically for seed yield and essential oil yield. The Medium Maturity Synthetic consistently displayed the highest average yield across both years, establishing itself as the most productive and stable genotype. In contrast, the Late Maturity Synthetic exhibited lower stability over the years. Consequently, the Medium Maturity Synthetic, showcasing superior yield and stability, emerges as a promising candidate for commercial release. These results highlight the importance of multi-environment field trials to understand genotype-environment interactions, especially for regions with variable climates. Further investigation of our genotypes across multiple locations could help more precisely identify stable genotypes for large-scale production.

Seed yield, as a complex trait, is influenced by several primary components. In this study, some variables including number of seeds per umbel, number of umbels, number of umbellets, number of seeds per umbellet, and essential oil yield, exhibited positive associations with seed yield. Earlier studies have consistently highlighted a strong and significant correlation between the number of umbels and seed yield in fennel (Zahid et al., 2008; Yadav et al., 2013; Sefidan et al., 2014). Additionally, Dwivedi et al. (2008) documented a robust and statistically significant association between seed yield and number of seeds per umbel. Furthermore, Bahmani et al. (2012) discovered significant connections between seed yield and variables such as the number of umbels, number of seeds per umbel, and stem diameter in their investigation. These meaningful associations underscore the importance of selecting genotypes characterized by higher numbers of umbels and seeds per umbel to enhance fennel yield. This emphasizes the necessity of improving these yield components to boost overall seed production in fennel.

4.2. Essential oil composition

Fennel essential oil primarily consists of three main groups of oxygenated monoterpenes, followed by hydrocarbon monoterpenes and hydrocarbon sesquiterpenes. Essential oil components such as *trans*-anethole, methyl chavicol, and fenchone, along with the main hydrocarbon monoterpene of limonene, contribute to the therapeutic properties of fennel (Osman and El-Wahab, 2009).

Trans-anethole varies across genotypes, with Fasa and the Medium Maturity Synthetic showing the highest percentages. Our finding about the essential oil compositions aligns with previous reports in Iranian fennel landraces, where *trans*-anethole ranged from 1.2 to 88.4%, methyl chavicol from 0.2 to 59.1%, fenchone from 1.1 to 14.7%, and limonene from 5.3 to 15.7% (Bahmani et al., 2016). Similar results were also reported from fennel populations in Yugoslavia (Mimica-Dukic et al., 2003) and Egypt (Viuda-Martos et al., 2010). The relative concentrations of these compounds exhibit significant variations based on the plant phenological stage and also the origin of the fennel. However, in our study, *trans*-anethole emerged as the primary component in essential oil, in contrast to a study in Turkey that reported a diverse range of oils from fennel seeds, where methyl chavicol was identified as the most prevalent component (Telci et al., 2019).

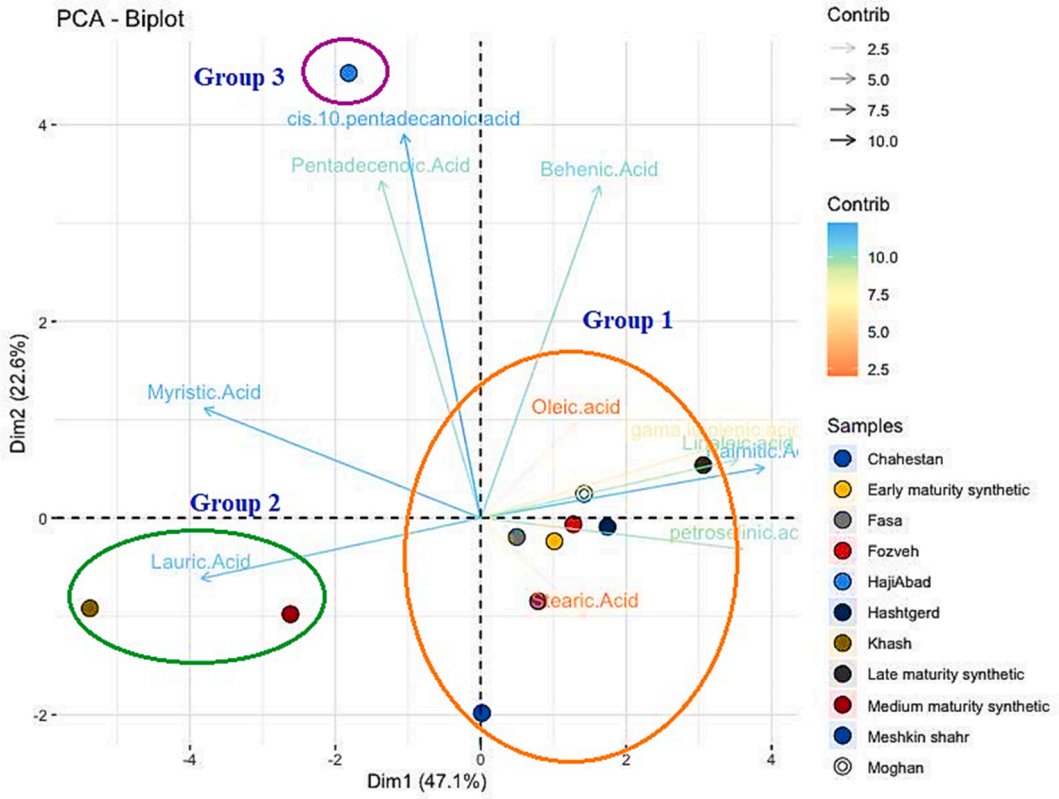


Fig. 6. Principle component analysis for main fatty acid profile of fennel genotypes.

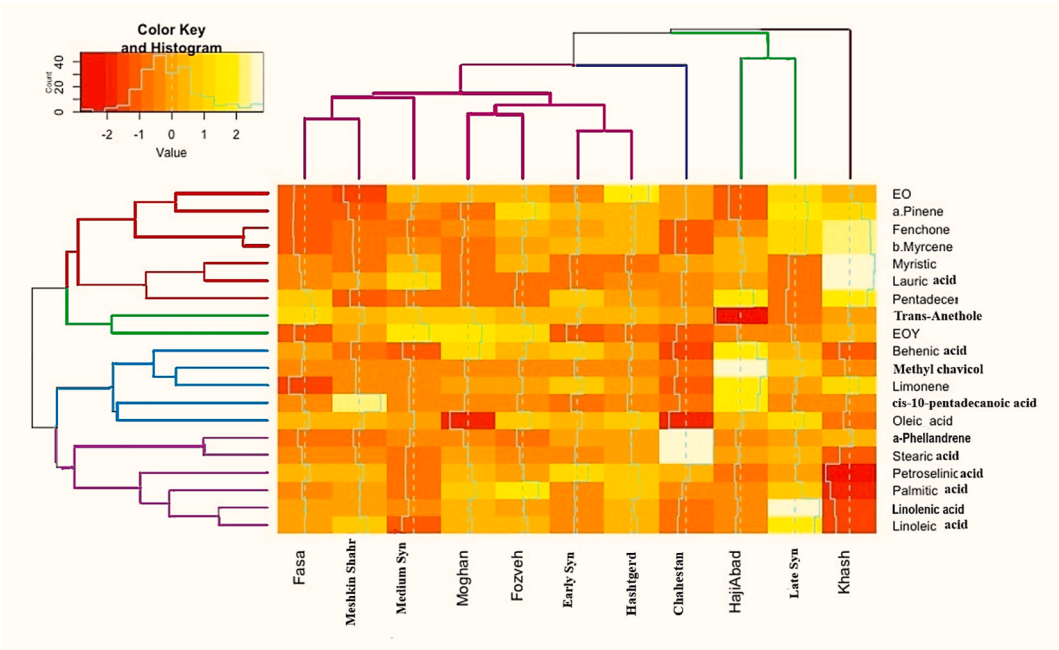


Fig. 7. Heat Map Analysis (HMA) depicting the primary essential oil components and fatty acid profiles across fennel genotypes.

Trans-anethole not only contributes to the delightful aroma and taste of fennel but is recognized for its antitumor characteristics through inducing apoptosis in cancer cells. Fennel cultivars with elevated *trans*-anethole content are particularly suitable for applications in the pharmaceutical and food industries (Rebey et al., 2016).

Fennel essential oil is considered of high quality when concentrations of methyl chavicol and fenchone are below 10%, imparting a sweet taste suitable for the food industry (Bilia et al., 2002; Bahmani et al., 2016). Among the three synthetic cultivars in our study, the Medium Maturity Synthetic and Early Maturity Synthetic cultivars exhibited low percentages of methyl chavicol and fenchone, remaining within favorable limits for these compounds. Additionally, they demonstrated a high content of *trans*-anethole, making them suitable for applications in the food industry and pharmaceuticals.

4.3. Fatty acid profiles

Fatty acids serve as significant sources of metabolic and energy storage, playing crucial roles in the early development and overall metabolism of humans (Rustan and Drevon, 2005). This study reveals that the composition of fatty acids is significantly influenced by the genotype. Petroselinic and linoleic acids emerged as the major unsaturated fatty acids in our study. The observed range of 60–78% petroselinic acid and range of 8.70–11.60% linoleic acids aligns with findings from previous fennel studies. For instance, Sayed Ahmad et al. (2017) reported petroselinic acid at 74.80%, along with linoleic acid at 12.74%, as the predominant fatty acids in fennel oil. Similarly, Najdoska-Bogdanov et al. (2015) indicated that petroselinic and oleic acids constituted the main fatty acids, ranging from 75.0% to 82.8%, with linoleic acid levels between 10.8% and 16.2%. Consistent with these findings, Bettaieb Rebey et al. (2016) reported petroselinic acid as the primary component at 75.43%, with linoleic acid at 8.04%. Yaldiz and Camlica (2019) also discovered petroselinic acid levels ranging from 49% to 87%. In our study, the Early Maturity Synthetic exhibited the highest percentage of petroselinic acid compared to the other genotypes. Beyond nutritional benefits, the richness of monounsaturated fatty acids, particularly petroselinic acid in fennel oil is an important factor in skincare and antiaging products. The potential application of fennel oil in skincare underscores its multifaceted utility beyond its nutritional value.

The highest amount of oleic acid was found in the Late Maturity Synthetic. The presence of oleic acid (omega 9) in food reduces arrhythmias and strokes and also helps lower blood pressure. Also, it lowers LDL cholesterol levels and increases HDL levels in the body. Late Maturity Synthetic showed the highest amount of omega 6. This acid is an essential fatty acid for the human body. This acid is vital to produce hormones such as prostaglandins, and effectively prevents blood clots in the arteries and inflammation of the arteries. In the case of polyunsaturated fatty acids, the amount of γ-linolenic acid (omega 3) in the Late Maturity Synthetic showed the highest amount of omega-3 fatty acids in humans have anti-tumor properties against various types of cancer. Overall, the synthetic cultivars demonstrated notable enrichment of beneficial unsaturated fatty acids like petroselinic and linoleic acid versus parental populations.

4.4. Multivariate analysis

Multivariate techniques, especially in metabolomics, are crucial for assessing diverse aspects including food quality, processing, and component analysis. (Cevallos-Cevallos et al., 2009; Wishart, 2008). In this study, heat maps and Principal Component Analysis (PCA) were employed to explore the compositions of essential oil and fatty acids in the synthetic fennel cultivars and their parental population. The PCA analysis categorized fennel genotypes into three groups based on major essential oil components: Group 1 (methyl chavicol chemotype), Group 2 (methyl chavicol/*trans*-anethole chemotype), and Group 3 (*trans*-anethole chemotype). This classification aligns with observations in Italian bitter fennels (Edoardo et al., 2010) and Iranian fennel landraces (Bahmani et al., 2016). The Medium Maturity Synthetic cultivars consistently showed itself as a *trans*-anethole chemotype, while Late Maturity Synthetic and Early Maturity Synthetic fell into Group 2 (methyl chavicol/*trans*-anethole chemotype). This finding corresponds with the study by Yaldiz and Camlica (2019), which employed PCA to categorize fennel genotypes into three groups based on essential oil components, revealing *trans*-anethole predominantly on the negative side of PC2 and methyl-chavicol on the positive side of PC1.

Furthermore, PCA analysis was conducted on the major fatty acid components of the fennel genotypes which classified the genotypes into three groups. The Late Maturity Synthetic and Early Maturity Synthetic were clustered in Group 1, characterized by a high percentage of petroselinic acid, while the Medium Maturity Synthetic fell into Group 2, showing a high percentage of lauric acid. Yaldiz and Camlica (2019) similarly employed PCA in their study, categorizing fennel genotypes based on fatty acid components. Additionally, a heatmap was generated to illustrate variations in both fatty acid and essential oil components among fennel genotypes. The positive correlation between methyl chavicol and limonene, noted by Raal et al. (2012) and Bahmani et al. (2016), further highlights an association between these compounds. Furthermore, correlations were observed between certain fatty acid and essential oil components, suggesting shared biosynthetic pathways or isomeric relationships (Bajalan et al., 2018). The chemical diversity present in our breeding populations suggests their potential for diverse bioactive applications, with lines rich in *trans*-anethole suitable for pharmaceutical uses and those enriched in petroselinic acid suitable for cosmetic purposes.

5. Conclusions

One of the key objectives of breeding programs is to introduce high-yielding genotypes with stable performance. The results of this study underscored the potential of synthetic cultivars as an efficient breeding approach for develop high-yielding and stable fennel cultivars that also maintain the genetic diversity within the cultivars. The presence of genetic diversity in synthetic cultivars equips the cultivars with resilience against variable and unpredicted growth conditions. In conclusion, our study demonstrated the effectiveness of the synthetic cultivar breeding method in fennel. The simultaneous improvement of several quality-related traits in fennel through synthetic cultivar breeding is evident. Synthetic fennel cultivars, with superior seed yield, high essential oil yield, elevated proportions of the beneficial essential oil compounds such as anethole, and unsaturated fatty acid compounds such as petroselinic

acid can make fennel able to contribute to food, pharmaceutical, and health industries even more than before. These populations can offer more stable performance in harsh and unpredicted environments, providing rich sources of beneficial metabolites in a wider range of growth conditions. Although a multi-location field trial is necessary to more precisely assess stability and adaptability of these synthetic cultivars under a variety of agro-climatic situations. A comprehensive examination of biosynthetic pathways that influence oil and fatty acid accumulation will provide more information on how to improve desired components. Exploring the interactions between genetics, environment, and metabolites can help fennel breeding attempts.

CRediT authorship contribution statement

Azam Akbari: Writing – review & editing, Writing – original draft, Methodology, Investigation. **Keivan Bahmani:** Resources, Methodology, Data curation. **Nesibe Ebru Kafkas:** Supervision, Resources, Methodology. **Omer Faruk Bilgin:** Software. **Tareq Hamijo:** Methodology, Investigation, Data curation. **Ali Izadi Darbandi:** Supervision. **Mohsen Farhadpour:** Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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