

Assessment of genetic diversity in biochemical and morphological traits in Iranian dill genotypes

Original Article

Abstract:

As a historical vegetable crop, dill (*Anethum graveolens* L.) is cultivated for food or herbal medicine. This research investigated the genetic variation of thirty dill (*Anethum graveolens* L.) genotypes collected from various regions of Iran. The trial was performed using a randomized block scheme utilizing four replicates, and some morphological and biochemical traits were measured. The range of plant fresh and dry weights was 5.39 g and 1.01 g per plant, respectively, with mean values of 3.75 g and 1.01 g, respectively. The essential oil content exhibited variation, with an average of 1.43%. Cluster analysis of traits indicated that Group I included seeds of umbels and umbellets, plant fresh and dry weight, seed yield, umbellets of umbel, and nodes before flowering, while Group II consisted of essential oil content and biochemical compounds such as proline, phenols, and anthocyanins. Group III included traits related to photosynthetic pigments, and Group IV consisted of fresh and dry weights of the inflorescence and the length of the flowering stem. Clustering of the dill genotypes grouped them into five clusters based on performance. Cluster I and Cluster II exhibited the highest values for plant dry weight and seed yield, indicating superior biomass production and reproductive capacity. Essential oil content was notably high in Cluster II and Cluster III, while Cluster IV and Cluster-V showed moderate to low levels in these target traits. Cluster II genotypes, encompassing eight accessions from diverse climatic regions, displayed high performance for seed yield, biomass, and essential oil content. The results highlight significant genetic variation within Iranian dill germplasm, emphasizing the potential for selecting superior genotypes had adaptability to a range of environmental circumstances. The identified high-performing genotypes, particularly those in Cluster II, are promising candidates for future breeding programs to enhance yield and quality traits.

Key words:

cluster analysis, cutoff line, essential oil content, multivariate analysis

Apstrakt:

Procena genetičke raznovrsnosti biohemijskih i morfoloških osobina kod iranskih genotipova mirođije

Kao tradicionalna povrtarska kultura, mirođija (*Anethum graveolens* L.) gaji se za ishranu ili kao lekovita biljka. Ovo istraživanje je ispitivalo genetičku varijabilnost trideset genotipova mirođije (*Anethum graveolens* L.) prikupljenih iz različitih regiona Irana. Ogled je izveden po randomizovanom blok sistemu sa četiri ponavljanja, pri čemu su merene određene morfološke i biohemijske osobine. Raspon sveže i suve mase biljaka iznosio je 5,39 g i 1,01 g po biljci, istim redom, sa prosečnim vrednostima od 3,75 g i 1,01 g. Sadržaj etarskog ulja pokazao je varijabilnost, sa prosekom od 1,43%. Klaster analiza osobina pokazala je da Grupa I obuhvata seme štitova i štitića, svežu i suhu masu biljke, prinos semena, broj štitića u štitu i broj nodusa pre cvetanja, dok je Grupa II obuhvatila sadržaj etarskog ulja i biohemijska jedinjenja kao što su prolin, fenoli i antocijanini. Grupa III uključivala je osobine povezane sa fotosintetskim pigmentima, dok je Grupa IV obuhvatila svežu i suhu masu cvasti i dužinu cvetne stabljike. Grupisanje genotipova mirođije svrstalo ih je u pet klastera na osnovu performansi. Klaster I i Klaster II pokazali su najviše vrednosti suve mase biljke i prinosa semena, što ukazuje na superiornu produkciju biomase i reproduktivni kapacitet. Sadržaj etarskog ulja bio je posebno visok u Klasteru II i Klasteru III, dok su Klaster IV i Klaster V pokazali umerene do niske vrednosti ovih ciljnih osobina. Genotipovi Klastera II, koji obuhvataju osam akcesija iz različitih klimatskih regiona, pokazali su visoke performanse u pogledu prinosa semena, biomase i sadržaja etarskog ulja. Rezultati ukazuju na značajnu genetičku varijabilnost unutar iranskog germplazma mirođije, naglašavajući potencijal za selekciju superiornih genotipova sa dobrom adaptabilnošću na različite uslove sredine. Identifikovani visokoperformantni genotipovi, posebno oni iz Klastera II, predstavljaju perspektivne kandidate za buduće oplemenjivačke programe usmerene na unapređenje prinosa i kvalitativnih osobina.

Ključne reči:

klaster analiza, granična linija, sadržaj etarskog ulja, multivarijantna analiza

Naser Sabaghnia

Department of Plant Production and Genetics,
Faculty of Agriculture, University of Maragheh,
5518779842, Maragheh, Iran
ORCID: 0000-0001-9690-6525
sabaghnia@maragheh.ac.ir (Corresponding author)

Mehdi Mohebodini

Department of Horticultural Science, Faculty
of Agricultural Science and Natural Resources,
University of Mohaghegh Ardabili, Ardabil, Iran
ORCID: 0000-0001-6359-3200

Asghar Ebadi

Department of Plant science, Moghan College of
Agriculture and Natural Resource, University of
Mohaghegh Ardabili, Ardabil, Iran
ORCID: 0000-0001-7132-7762

Mohsen Janmohammadi

Department of Plant Production and Genetics,
Faculty of Agriculture, University of Maragheh,
5518779842, Maragheh, Iran
ORCID: 0000-0002-6121-6791

Received: February 14, 2026

Revised: February 20, 2026

Accepted: February 25, 2026



Introduction

Plant germplasm represents one of the most critical and valuable natural resources, with the primary objectives of its management being collection, identification, and long-term conservation for future generations. Medicinal and vegetable crops, such as dill (*Anethum graveolens* L.), are of considerable economic importance and are widely used in both fresh and processed forms (Lal et al., 2023). A comprehensive understanding of genetic diversity within plant species is essential for the effective selection of parent lines in hybridization programs, ultimately contributing to the development of improved cultivars. Dill, a dual-purpose plant serving as both a culinary herb and a medicinal species, is believed to have originated in Turkey and Southern Europe (Yaldız et al., 2018). It also grows wild across various regions, including the Caucasus, northeastern Africa, and Iran (Wahba et al., 2025). According to historical accounts, systematic cultivation of dill for its aromatic and therapeutic qualities began in the Middle East as early as the 14th century and gradually spread to other regions of the world. In recent decades, global dill production has shown a notable upward trend. According to FAOSTAT (2020), worldwide dill production has reached approximately 200,000 tons, with leading producer countries including India, Iran, Turkey, and Egypt. This increased demand highlights the significance of dill not only as a cultural and culinary staple but also as a valuable genetic resource for future breeding and conservation efforts.

Dill is characterized by a straight, conical, white-colored root and a cylindrical, glabrous stem with longitudinal ridges and slight depressions at the nodes (Mohebodini & Farmanpour-Kalalagh, 2021). The fruit is elliptical and flattened, measuring approximately 4–5 mm in length and about 3 mm in width, with a light chocolate-brown coloration. These fruits are widely recognized for their medicinal properties (Ebadi et al., 2025). Primary umbels, bearing golden-yellow flowers, emerge from the terminal portion of the main stem, while secondary umbels develop along the stem throughout the growing season (Gholizadeh et al., 2021). Both fresh and dried aerial parts, as well as dill seeds, are used in traditional and modern medicinal practices. Because of its antibacterial properties, dill essential oil is frequently used to treat gastrointestinal issues such as bloating and stomachaches. It has also been shown to lower blood cholesterol levels (Mohammed et al., 2019). Essential oils are present in all aerial parts of the plant, although their concentration varies among different organs. In vegetative tissues, the major constituents of essential oil (0.8–1.6%) are D-carvone

and α -phellandrene (Najaran et al., 2016). Assessing genetic diversity among plant populations is a crucial preliminary step in identifying, sustainably utilizing, and effectively conserving genetic resources, which are vital to genetic research and breeding programs. Without adequate genetic variability, plant breeders face significant limitations in developing new and improved cultivars. Understanding the extent and inheritance patterns of phenotypic and genotypic variation within plant ecotypes provides a solid foundation for selecting suitable parental lines in breeding initiatives aimed at producing superior genotypes in future generations.

Solouki et al. (2012) examined genetic variation among 37 dill genotypes using both morphological and molecular markers. Their findings revealed strong associations among phenological traits, and cluster analysis categorized the genotypes into four distinct groups. Furthermore, principal component analysis identified 13 components that collectively explained approximately 70% of the dataset's total variability. Similarly, Ninou et al. (2017) evaluated genetic variation among 33 dill genotypes and clustered them into three geographical groups: the northern region, the Aegean region, and central Greece. They reported considerable variation in yield-related traits, such as leaf number and umbel diameter, which can be exploited in plant improvement projects to enhance plant biomass and seed production. Kadoglidou et al. (2023) evaluated 22 genotypes and 9 commercial varieties of dill, identifying several promising genotypes suitable for genetic enhancement. These genotypes showed potential to develop improved varieties with favorable agronomic traits, including higher aromatic content, increased yield, early maturity, and enhanced nutritional quality. The study also emphasized the significance of combining phytochemical and agronomic characteristics as useful instruments for plant enhancement initiatives. Maintaining agricultural development initiatives requires the gathering, preservation, and methodical assessment of genetic resources. In this context, the current study was conducted to assess genetic variation among dill genotypes for key morphological traits and economically valuable characteristics. The primary objective was to identify superior accessions that can serve as parental lines in future breeding initiatives to improve dill's agronomic and commercial value.

Materials and Methods

Trail and morphological traits

In the present study, seeds from thirty dill genotypes originating from various regions across Iran, representing considerable geographical

diversity, were collected and evaluated (Tab. 1). The experiment was conducted in real-world conditions employing a randomized block design with four replicates. Each plot measured 80 × 60 cm, and the seeds were planted by hand. Seedling emergence occurred within two to three weeks for all genotypes, while standard agronomic practices, including regular irrigation and manual weed control, were applied throughout the growing season.

Approximately three weeks after the appearance of the first flowers, when 50% flowering was observed, morphological evaluations were performed. For each plot, seven plants were randomly selected from the front, middle, and rear sections of the rows to ensure sampling uniformity. The following morphological traits were recorded: flowering stem length (FSL), number of umbels per plant (NUP), number of nodes before flowering (NNF), number of seeds per umbel

Table 1. Name and some geographical attributes of dill genotypes

Code	Name	Province	Longitude	Latitude	Altitude
G1	Jowkar	Hamedan	48°41'N	34°25'E	1694
G2	Andimeshk	Khuzestan	48°21'N	32°27'E	176
G3	Malayer	Hamedan	48°49'N	34°17'E	1746
G4	Meshkinshar-A	Ardabil	47°40'N	38°23'E	1400
G5	Tabriz	East-Azarbaijan	47°40'N	38°23'E	1348
G6	Jahrom	Fars	53°33'N	28°30'E	1050
G7	Eslamshahr	Tehran	51°13'N	35°32'E	1020
G8	Hamedan-A	Hamedan	48°30'N	34°47'E	1850
G9	Ardabil	Ardabil	38°15'N	48°17'E	1351
G10	Ahvaz	Khuzestan	48°43'N	31°20'E	16
G11	Sarab	East-Azarbaijan	47°32'N	37°56'E	1683
G12	Varamin	Tehran	51°38'N	35°19'E	920
G13	Meshkinshar-B	Ardabil	46°18'N	38°40'E	1418
G14	Tehran	Tehran	51°22'N	35°42'E	1214
G15	Damghan	Semnan	54°20'N	36°90'E	1154
G16	Kerman	Kerman	57°40'N	30°17'E	1767
G17	Jolfa	East-Azarbaijan	45°37'N	38°56'E	707
G18	Mashhad	Razavi-Khorasan	59°32'N	36°19'E	1027
G19	Esfahan	Esfahan	51°40'N	32°40'E	1575
G20	Gorgan	Golestan	54°25'N	36°50'E	133
G21	Kashmar	Razavi-Khorasan	58°27'N	35°14'E	1051
G22	Urmia	West-Azarbaijan	45°40'N	37°33'E	1348
G23	Shiraz	Fars	52°32'N	29°35'E	1519
G24	Astara	Gilan	48°52'N	38°25'E	-24
G25	SomehSara	Gilan	49°18'N	37°18'E	6
G26	Dezful	Khuzestan	48°24'N	32°23'E	142
G27	Gazvin	Gazvin	50°00'N	36°16'E	1310
G28	Bushehr	Bushehr	50°50'N	28°57'E	11
G29	Rezvanshahr	Gilan	51°50'N	32°42'E	1889
G30	Hamedan-B	Hamedan	48°30'N	34°47'E	1818

(SNU), number of seeds per umbellet (NSU), and number of umbellets per umbel (NUU). In addition, several biomass-related traits were measured, including inflorescence fresh weight (IFW), plant fresh weight (PFW), inflorescence dry weight (IDW), and plant dry weight (PDW). All weights were determined using a precision balance with ± 0.01 g accuracy. To ensure accurate measurement of dry-related traits, samples were dried in an oven at 70 °C for one day. Seed yield (SY) was recorded at the time of full maturity.

Biochemical traits

To determine the essential oil content (EOC), shade-dried samples were hydrodistilled using a Clevenger apparatus. Specifically, a 50 g sample was distilled with 500 mL of water for three hours at 28 °C. The collected essential oil was then measured and expressed as a percentage of the dry plant weight. Photosynthetic pigments, including anthocyanin (Ant), chlorophyll a (CHLa), carotenoids (Car), and chlorophyll b (CHLb), were quantified following the protocol described by Ferreira-Barreto et al. (2018). Fresh tissue samples (200 mg) were homogenized in 2.0 mL of an extraction solution composed of 85% acetone and 15% Tris buffer (1% w/v). After centrifugation at $12,000 \times g$ for 3 minutes, 1.0 mL of the supernatant was diluted to 3.0 mL. Absorbance readings were taken at 537, 663, 647, and 470 nm using a spectrophotometer to quantify the pigment concentrations. Total phenol content (Phe) was estimated according to Sarker et al. (2018). Fresh plant tissue (50 mg) is homogenized in 80% acetone and centrifuged at $10,000 \times g$ for 10 minutes. Then, 100 μ L of the resulting sample was mixed with 2 mL of distilled water and 1 mL of Folin-Ciocalteu reagent. Then it was thoroughly shaken, and the phenol content was quantified spectrophotometrically. Proline (Pro) content was measured following the method of Ikram et al. (2018). For this, a 0.5 g sample was homogenized and centrifuged at $4,000 \times g$ for 10 min at 4 °C with 3% sulfosalicylic acid. The sample obtained was treated with acid ninhydrin and glacial acetic acid, and was incubated at 98 °C for 30 min. After cooling, the chromophore was extracted with toluene, and absorbance was measured at 520 nm to determine the concentration of proline.

Data analysis

To ensure the assumptions of parametric analyses were met, the homogeneity of residuals was examined using the Levene test, and normality was assessed using the Kolmogorov-Smirnov test. Hierarchical agglomerative clustering was employed to group both the dill genotypes and their associated traits. This was accomplished

using squared Euclidean distances calculated from standardized data, which provided a measure of dissimilarity between observations. The formation of a cluster diagram was performed via the minimum variance procedure of Ward, which merges clusters by minimizing the increase in total within-Cluster-Variance. These analyses were performed using Statistica software version 14.0 (TIBCO Inc., USA). To statistically validate the clustering outcomes, multivariate analysis of variance (MANOVA) was performed. Four multivariate test criteria: largest root of Roy, Wilks' lambda, Pillai's trace, and Hotelling-Lawley trace, and Roy's largest root, were applied to determine the statistical significance of differences among the identified clusters. These tests evaluated the extent to which genotypic groups differed across the measured traits. The number of significant clusters was established by interpreting these multivariate statistics, thereby ensuring the selected cluster solution captured the maximum genetic divergence within the dill germplasm. All multivariate parameters were computed using SPSS 27.0 (SPSS Inc., USA).

Results

Descriptive statistics

The range of plant fresh weight (PFW) and plant dry weight (PDW) among the dill genotypes was 5.39 g and 1.01 g per plant, respectively, with corresponding mean values of 3.75 g and 1.01 g per plant (**Tab. 2**). The essential oil content (EOC) also exhibited variation, ranging up to 1.43%, with an average value of 1.43%. The coefficient of variation (CV) was used to assess the extent of variability among traits and was classified as low (<20%), moderate (20–30%), or high (>30%). Among the measured traits, phenol content (Phe) exhibited the lowest CV, indicating limited variability and suggesting a more conserved expression across genotypes (**Tab. 2**). Traits showing moderate levels of variability included number of nodes before flowering (NNF), number of umbellets per umbel (NUU), number of seeds per umbellet (NSU), carotenoid content (Car), chlorophyll b (CHLb), seed yield (SY), EOC, and PDW; pointing to an intermediate degree of phenotypic divergence. Traits such as anthocyanin (Ant), proline (Pro), flowering stem length (FSL), number of seeds per umbel (SNU), inflorescence fresh weight (IFW), number of umbels per plant (NUP), chlorophyll a (CHLa), inflorescence dry weight (IDW), and PFW displayed high CV values, indicating considerable variation among the studied accessions. These results underscore substantial phenotypic diversity among the evaluated dill genotypes, particularly in traits related to yield, such as SNU and NUP. Such

Table 2. Descriptive parameters of dill traits.

Traits	Mean	SE	CV	Range
NSU	7.98	0.3	20.6	6.43
SNU	158.3	11.7	40.5	250.3
NUU	13.21	0.63	26.3	13.05
NUP	2.72	0.19	38.1	4.14
NNF	4.99	0.19	20.8	3.68
FSL	5.64	0.33	32.1	6.92
EOC	1.43	0.07	25.3	1.43
Car	0.41	0.02	28.6	0.52
CHLa	0.64	0.03	29.7	0.7
CHLb	2.04	0.12	32.2	2.63
Phe	15.78	0.46	15.9	11.01
Pro	4.42	0.35	43.3	7.37
Ant	11.38	0.7	33.6	13.42
IFW	0.52	0.04	38.4	0.83
PFW	3.75	0.27	38.8	5.39
IDW	0.14	0.01	39.5	0.21
PDW	1.01	0.04	20.6	1.01
SY	0.49	0.02	23.5	0.46

Traits are number of seed per umbellet (NSU), seed number per umbel (SNU), number of umbellet per umbel (NUU), number of umbels per plant (NUP), node number before flowering (NNF), flowering stem length (FSL), essential oil content (EOC), carotenoid (Car), chlorophyll b (CHLa), chlorophyll a (CHLb), phenol (Phe), proline (Pro), anthocyanin (Ant), seed yield (SY), inflorescence fresh weight (IFW), plant fresh weight (PFW), inflorescence dry weight (IDW), and plant dry weight (PDW).

variation is of significant importance to breeding programs, as it provides opportunities to identify and select superior genotypes with enhanced seed yield and biomass productivity. Similar patterns of variability in dill have been previously reported by Mohebodini and Farmanpour-Kalalagh (2021), who observed pronounced CV values for yield components and biomass traits across various dill genotypes.

Clustering of traits

The trait dendrogram (Fig. 1) was employed to explore patterns of interrelationships among the

measured variables. To validate the optimal number of trait clusters, multivariate analysis of variance was conducted using four robust test statistics: the Roy's largest root, Wilks' lambda, and the Pillai's and Hotelling-Lawley traces (Tab. 3). All these statistical indicators were highly significant, confirming the partitioning of traits into four distinct clusters. Group I included number of seeds per umbellet (NSU), plant dry weight (PDW), number of seeds per umbel (SNU), seed yield (SY), number of umbellets per umbel (NUU), plant fresh weight (PFW), and number of nodes before flowering (NNF). This grouping indicates a strong interrelationship between seed yield and biomass traits with structural components of the plant, particularly the architecture of the inflorescence (umbels and umbellets) and developmental traits such as node number. The multivariate analysis and trait clustering identify key relationships among plant traits, including seed yield, biomass, and inflorescence structure. This information is valuable for optimizing seed yield by selecting for traits linked to biomass and plant architecture, improving crop breeding by targeting traits that enhance both reproductive and structural performance, and refining crop management practices, such as plant spacing and pruning, to maximize productivity. Additionally, the analysis helps predict crop performance under varying environmental conditions and guides resource allocation, ensuring that inputs are directed towards the most influential traits for yield improvement. Ultimately, it supports the development of more productive, efficient, and sustainable crop varieties. The clustering suggests that these traits collectively enhance dill's yield potential, supporting the hypothesis that both seed and biomass yields are influenced by multiple, interacting morphological components. These findings are consistent with those of Sinhasane et al. (2022), who reported that yield traits in dill are governed by complex, often non-linear interactions among several structural traits. Group II consisted of essential oil content (EOC) and biochemical compounds such as proline (Pro), phenols (Phe), and anthocyanins (Ant). The grouping of these variables reflects their common involvement in the plant's secondary metabolism, which is often associated with stress responses and medicinal properties. The results align with those of Dehghani et al. (2024), who demonstrated significant positive associations between essential oil content and these biochemical traits in dill. Group III included traits related to photosynthetic pigments, namely chlorophyll a (CHLa), carotenoids (Car), and chlorophyll b (CHLb). This cluster highlights the biochemical coherence among pigments involved in light harvesting and photosynthesis.

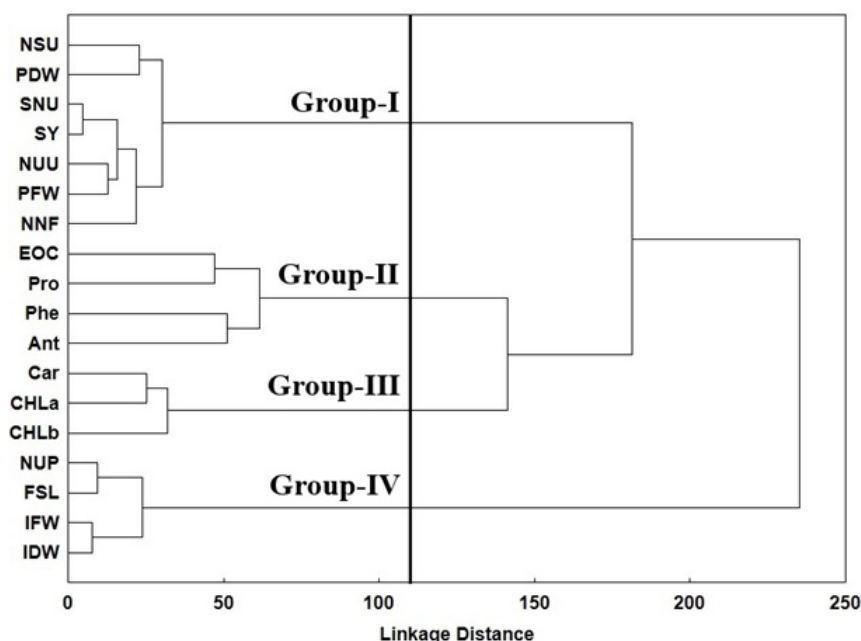


Fig. 1. The diagram of cluster analysis for grouping traits of dill. Traits are number of seed per umbellet (NSU), seed number per umbel (SNU), number of umbellet per umbel (NUU), number of umbels per plant (NUP), node number before flowering (NNF), flowering stem length (FSL), essential oil content (EOC), carotenoid (Car), chlorophyll b (CHLb), chlorophyll a (CHLa), phenol (Phe), proline (Pro), anthocyanin (Ant), seed yield (SY), inflorescence fresh weight (IFW), plant fresh weight (PFW), inflorescence dry weight (IDW), and plant dry weight (PDW)

Similar associations were reported by Majdoub et al. (2017), who found strong correlations between carotenoids and chlorophyll content in dill. These results collectively underscore the complexity of yield determination in dill, in which performance is not merely the sum of linear additive components but rather the outcome of dynamic interactions among physiological, morphological, and biochemical traits. Group IV comprised FSL, IFW and IDW, indicating the close relation of flowering traits (fresh and dry weights of inflorescence and length of flowering stem), with each other. In many crops, yield is frequently modeled as the linear product of its components; however, emerging evidence suggests it is also shaped by factors such as sink capacity during reproductive development and source activity during vegetative growth (Smith et

al., 2018). Therefore, a deeper physiological understanding of the interplay between vegetative and reproductive phases is essential for guiding breeding efforts to enhance yield. Based on current findings, selecting genotypes with a high number of umbels, umbellets, and seeds per umbel is likely to improve both seed yield and biomass accumulation. This reinforces the multifaceted nature of yield as a trait in dill and highlights the importance of incorporating multi-trait interactions into future breeding strategies.

Clustering of genotypes

A second dendrogram was constructed to investigate the clustering patterns among the thirty dill genotypes (Fig. 2). The determination of the optimal cutoff line for cluster delineation was validated using multivariate test statistics, including Roy's largest root, Wilks' lambda, and the trace criteria of Pillai and Hotelling, all of which showed significant values (Tab. 4). Based on these criteria, the genotypes were grouped into five distinct clusters. The mean trait values for each cluster are summarized in Tab. 5, providing insight into the defining characteristics of each group. Cluster I comprised a single genotype, G24 (Astara), which exhibited the highest mean values for nearly all measured traits, except for chlorophyll a (CHLa), where it showed a moderate level, and for essential oil content (EOC), carotenoids (Car), chlorophyll b (CHLb), and proline (Pro), where values were comparatively low. Cluster II included eight genotypes characterized by the highest means across most traits, with moderate levels of seed number per umbel (SNU), phenols (Phe), and inflorescence dry weight (IDW), but comparatively lower values

Table 3. Multivariate analysis of variance for determining the cutoff line in traits

Parameter	Value	F	Hypothesis df	Error df	Sig.
Pillai's Trace	2.98	40.6	42	9	0
Wilks' Lambda	0.00086	51.68	42	3.7	0
Hotelling-Lawley's Trace	7102.21	591.85	42	12	0
Roy's Largest Root	4868.15	1043.18	14	3	0

for number of umbels per plant (NUP), flowering stem length (FSL), and inflorescence fresh weight (IFW). Cluster III comprised four genotypes (G4, G8, G12, and G20), which were distinguished by high mean values primarily for essential oil content (EOC) and number of nodes before flowering (NNF). Cluster IV contained twelve genotypes exhibiting elevated anthocyanin (Ant) concentrations, coupled with lower plant dry weight and moderate values for other traits, including essential oil content. Cluster-V was made up of five genotypes (G1, G2, G3, G14, and G15) having the highest accumulation of photosynthetic pigment (Car, CHLa, and CHLb) as well as moderate values of most yield components and essential oil content. These distinct clusters demonstrate substantial variability among genotypes for key agronomic, biochemical, and morphological traits, underscoring their potential as valuable genetic resources. The diversity observed across clusters offers promising opportunities for targeted breeding programs aimed at specific goals, such as improving seed yield, biomass production, or enhancing essential oil content. This clustering analysis highlights the feasibility of selecting and advancing genotypes with desirable trait combinations for future cultivar development and genetic improvement in dill genotypes.

Regarding key target traits such as plant dry weight, genotypes in Cluster I and Cluster II exhibited the highest values, while those in Cluster III and Cluster V showed moderate levels, and genotypes in Cluster IV displayed the lowest values. In terms of essential oil content, genotypes within Cluster II and Cluster III recorded the highest concentrations, whereas Clusters IV and V had moderate amounts, and Cluster I genotypes demonstrated comparatively

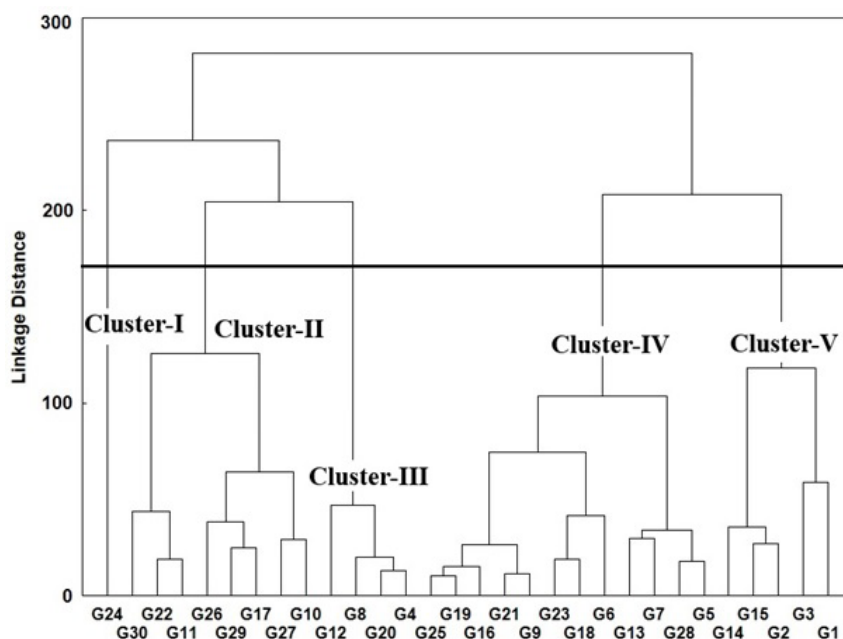


Fig. 2. The diagram of cluster analysis for grouping 30 dill genotypes

low essential oil levels. When evaluating seed yield performance, Clusters I and II again showed superior results, Cluster III and IV presented moderate yields, and Cluster V had the lowest seed yield among the groups. Based on these evaluations, the genotypes within Cluster II appear to be the most suitable candidates in terms of combined target traits: biomass, seed yield, and essential oil content, and thus represent promising material for cultivation and potential variety release. The genotypes comprising Cluster II were G10 (Ahvaz), G11 (Sarab), G17 (Jolfa), G22 (Urmia), G26 (Dezful), G27 (Qazvin), G29 (Rezvanshahr), and G30 (Hamedan-B). These genotypes originate from diverse climatic regions across Iran. However, genotypes G11, G17, G22, and G27 come from the northwest, characterized by a cool climate; G10 and G26 are from the southwest, an area with warmer conditions; G29 is from the humid northern region; and G30 hails from the semi-arid, cool western area. This broad geographic representation suggests that the superior genotypes identified in Cluster II are well-adapted to a wide range of environmental conditions, making them highly suitable for successful cultivation by local

Table 4. Multivariate analysis of variance for determining the cutoff line in genotypes

Parameter	Value	F	Hypothesis df	Error df	Sig.
Pillai's Trace	3.44	3.78	72	44	0
Wilks' Lambda	0.001	4.5	72	33.8	0
Hotelling-Lawley's Trace	55.8	5.04	72	26	0
Roy's Largest Root	36.05	22.03	18	11	0

Table 5. The means of traits for five identified clusters of dill genotypes

Traits	Cluster-I	Cluster-II	Cluster-III	Cluster-IV	Cluster-V
NSU	10.57	9.27	7.4	6.74	8.83
SNU	333	221.3	151	112.2	139.2
NUU	20.66	17.21	13.41	11.04	10.4
NUP	5.18	2.02	1.34	3.38	2.85
NNF	5.66	5.96	5.95	4.14	4.57
FSL	8.36	4.19	3.8	6.6	6.58
EOC	1.01	1.62	1.51	1.32	1.42
Car	0.21	0.46	0.41	0.34	0.54
CHLa	0.68	0.75	0.58	0.49	0.86
CHLb	0.76	2.68	1.86	1.67	2.32
Phe	20.3	16.95	14.33	15.11	15.8
Pro	3.17	5.66	4.49	4.03	3.58
Ant	13.39	12.66	7.1	12.34	10.07
IFW	1.06	0.46	0.28	0.56	0.61
PFW	5.47	5.4	3.94	2.69	3.16
IDW	0.25	0.12	0.06	0.14	0.17
PDW	1.3	1.13	1.03	0.87	1.08
SY	0.71	0.6	0.51	0.4	0.46

Traits are number of seed per umbellet (NSU), seed number per umbel (SNU), number of umbellet per umbel (NUU), number of umbels per plant (NUP), node number before flowering (NNF), flowering stem length (FSL), essential oil content (EOC), carotenoid (Car), chlorophyll b (CHLa), chlorophyll a (CHLb), phenol (Phe), proline (Pro), anthocyanin (Ant), seed yield (SY), inflorescence fresh weight (IFW), plant fresh weight (PFW), inflorescence dry weight (IDW), and plant dry weight (PDW)

farmers across varied climatic zones.

Discussion

Cluster analysis provided valuable insights into the variability and relationships among important traits, such as seed yield, biomass, essential oil content, and photosynthetic pigments, which are highly significant for future dill breeding and cultivation strategies. The observed wide ranges and coefficients of variation in target traits, such as seed yield, plant fresh and dry weight, and essential oil content, reflect relatively high genetic diversity within the studied germplasm. Also, traits related to yield components (e.g., seeds per umbel and umbels per plant) showed high variability, highlighting a valuable genetic resource for selection and improvement. The practical application of this cluster analysis for dill breeding and cultivation involves leveraging the genetic diversity identified in key traits such as seed yield, biomass, and essential oil content. Breeders can use this diversity to select superior genotypes with desired traits,

thereby improving crop productivity and quality. The high variability in traits such as seeds per umbel and umbels per plant provides valuable targets for yield improvement and increased oil content. This insight also enables the optimization of cultivation practices, helping farmers adjust growing conditions, such as nutrient management and irrigation, to maximize yields and essential oil quality. Also, by focusing on highly variable traits, resources can be allocated more effectively to achieve higher overall productivity. Ultimately, this analysis supports the development of high-yielding, quality-focused dill varieties, benefiting both breeders and farmers by enhancing crop performance. Such extensive variation is consistent with previous studies on dill germplasm, like considerable variability in biomass and yield components in dill genotypes (Sinhasane et al., 2022). Similarly, Palumbo et al. (2021) found wide ranges in agronomic and phytochemical traits among dill accessions, emphasizing the presence of exploitable diversity for breeding purposes. These congruent results affirm that Iranian dill populations

harbor significant genetic potential, likely shaped by their adaptation to diverse climatic zones and local cultivation practices.

Hierarchical clustering and multivariate analyses categorized traits into four distinct groups, revealing meaningful biological relationships. Group I, including seed yield, biomass-related traits, and yield components, showed strong intercorrelations. This suggests that increasing umbel and seed numbers could directly enhance seed yield and biomass production. This finding corroborates the notion that dill yield is a complex trait influenced by multiple components that act in a non-linear manner, as emphasized by Biesiada et al. (2019). Group II, which includes essential oil content and biochemical markers such as proline, phenols, and anthocyanins, highlights a strong relationship among these traits. The co-clustering of these biochemical markers with essential oil content reinforces findings from Khan et al. (2020), which showed positive associations between essential oil yield and stress-related metabolites in dill. This suggests that dill plants with higher levels of stress-related metabolites may produce more essential oil, making them valuable for breeding and cultivation strategies to increase essential oil yield. Additionally, understanding the biochemical pathways involved can help optimize conditions that promote both stress resistance and oil production. According to this relationship, plants with higher levels of osmoprotectant and antioxidant chemicals may be able to sustain or increase the production of secondary metabolites under stress, providing both medicinal and aromatic quality benefits. Group III comprised photosynthetic pigments, reflecting the fundamental physiological processes governing biomass accumulation and photosynthetic efficiency. This cluster aligns with Wasli et al. (2018), who reported strong correlations among photosynthetic pigments in dill, indicating their co-regulation and collective role in energy capture and stress resilience. The trait grouping highlights the multifaceted nature of agronomic performance in dill, where physiological, biochemical, and morphological attributes interact intricately. Understanding these interrelationships enables breeders to target multiple traits simultaneously, improving yield and quality in an integrated manner.

The cluster analysis of dill genotypes revealed five distinct groups with contrasting trait profiles, reflecting adaptation to diverse agroclimatic regions and genetic backgrounds. Clusters I and II included genotypes with superior biomass and seed yield, making them prime candidates for cultivation and breeding programs focused on productivity. Notably, genotypes in Cluster II also exhibited high essential

oil content, combining yield and quality traits, a rare but desirable combination. The geographical origins of Cluster II genotypes highlight the significant role of environmental conditions in shaping genetic expression. The inclusion of accessions from diverse climates, ranging from cool (northwest Iran) to warm (southwest Iran) and humid (north Iran) to semi-arid (west Iran) regions, demonstrates that these genotypes have broad adaptability. This adaptability is a key trait for varietal release in rainfed and marginal areas, where environmental conditions can be unpredictable. Their resilience across various climates provides farmers with reliable, robust options for local cultivation, ensuring consistent production and income, even in challenging growing environments. High essential oil concentration and moderate yield were observed in Cluster III genotypes, suggesting their potential for breeding programs that prioritize aromatic quality over bulk production. The Cluster II genotypes could be crossed with high-yielding lines to combine both yield potential and adaptability in their progeny. This breeding approach would aim to develop plants that not only produce higher yields but are also resilient in diverse environmental conditions. Meanwhile, Cluster IV and Cluster V, characterized by moderate to low biomass and seed yield but possessing distinct physiological or biochemical profiles, highlight valuable niche germplasm. These genotypes may be useful for specific breeding objectives, such as stress tolerance or enhancement of phytochemicals (e.g., essential oils, antioxidants). By focusing on these traits, breeders can develop varieties with enhanced resilience or increased concentrations of beneficial compounds, even if they have lower yield potential. The intricate associations between yield components, biochemical traits, and physiological markers reveal that breeding efforts should adopt a multi-trait selection strategy. Focusing solely on seed yield may overlook valuable biochemical and physiological traits that confer stress tolerance and essential oil quality (Reis et al., 2026). The positive correlation of essential oil content with proline, phenols, and anthocyanins suggests that breeding for stress resilience can concurrently improve medicinal and aromatic properties. Given the increasing global demand for natural products with health benefits, enhancing these traits can add economic value to dill cultivars. Furthermore, clustering genotypes by diverse traits provides a roadmap for parent selection in hybridization programs. Combining high-yielding genotypes from Clusters I and II with aromatic-rich lines from Cluster III can generate progenies with balanced traits, addressing both productivity and quality. The study also highlights the importance of local adaptation. The identification of superior

genotypes across varied climatic zones supports the development of region-specific cultivars that are more likely to perform consistently under local environmental stresses, including drought and temperature extremes. Future research should also explore the genetic basis of essential oil biosynthesis pathways in dill, aiming to manipulate them through conventional or biotechnological means to enhance aromatic profiles.

Conclusion

The evaluated dill genotypes exhibited a wide range of desirable characteristics, making them valuable candidates for inclusion in future breeding efforts or direct release as improved cultivars. Among these, eight genotypes originating from Ahvaz, Sarab, Jolfa, Urmia, Dezful, Gazvin, Rezvanshahr, and Hamedan-B demonstrated superior performance in key traits, including biomass production, seed yield, and essential oil content.

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