

## Cluster Analysis of Genetic Diversity in Some Agronomic Traits Among Black Cumin (*Nigella sativa* L.) Landraces

Naser Sabaghnia<sup>1\*</sup>, Mahdi Mohebodini<sup>2</sup>, Asghar Ebadi<sup>1</sup>, Mohsen Janmohammadi<sup>1</sup>

Received: 2024-11-10 Accepted: 2025-01-11

### Abstract

Black cumin (*Nigella sativa* L.), a widely used medicinal and aromatic plant, has gained significant attention due to its diverse applications in the drug and food manufacturing. The current study assessed the genetic variation and agronomic performance of twenty-seven black cumin genotypes originating from various regions of Iran, focusing on a range of morphological traits and yield components. The genotypes were grown under controlled greenhouse conditions in a randomized complete block design with four replicates. Some traits, including plant height, flowering stem length, seed yield, capsule weight, and thousand-seed weight, were measured for each genotype. Statistical analysis revealed high genetic variation across most traits, with the coefficient of variation (CV) exceeding 20% for many characters, indicating significant potential for selection in breeding programs. The high variations of seed yield, seeds per capsule, and seed size suggested that these traits could be targeted for improvement in future breeding efforts. Agglomerative hierarchical cluster analysis was performed using standardized squared Euclidean distances to group the genotypes based on their genetic similarity. The resulting dendrogram identified four distinct clusters, with Cluster-A containing genotypes (G21, G23, G24, and G26) exhibiting the highest seed yield and overall favorable agronomic traits, making them prime candidates for breeding programs focused on high yield. Cluster-B included genotypes with large seed size and thousand-seed weight but lower overall yield performance. Cluster-C genotypes displayed a balance of high seed yield, seed size, and other yield components. Although Cluster-D performing poorly in terms of seed yield, contained genotypes with large seed size, which could be valuable for market-oriented breeding. The cluster analysis groups black cumin genotypes based on similarities in traits, such as seed size, oil content, or disease resistance, helping identify distinct genetic clusters. This information enables researchers and farmers to select superior genotypes from diverse clusters for breeding programs, ensuring the combination of complementary traits to improve crop yield, enhance stress tolerance, and increase overall productivity. However, four genotypes including Kashmar, Mashhad-I, Mashhad-II, and Miandoab, are recommended for cultivation through farmers and entering in breeding programs due to their high seed yield performance.

**Keywords** Dendrogram, Genetic variation, multivariate ANOVA

1-Department of Plant Production and Genetics, Faculty of Agriculture, University of Maragheh, Maragheh, Iran.

2-Department of Horticultural Science, Faculty of Agricultural Science and Natural Resources, University of Mohaghegh, Mohaghegh, Iran

\*The corresponding author email address:sabaghnia@yahoo.com

Doi: 10.48308/pae.2025.238366.1100



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## Introduction

Black cumin (*Nigella sativa* L.) from the Ranunculaceae family is an annual plant that produces blossoms and is native to the Mediterranean, North Africa, and Southwest Asia regions (Khadim et al., 2024). In Iran, it thrives in various regions, including Ardabil, Arak, Isfahan, Lorestan, Kermanshah, and Khorasan, where it is widely cultivated (Ghorbanzadeh-Neghab and Zare-Mehrjerdi, 2018; Mehri et al., 2022). Known for its numerous benefits, black cumin helps lower blood pressure, aids digestion, exhibits antibacterial properties, fights cancer, and relieves stress (Golkar and Nourbakhsh, 2019). It also supports respiratory and nervous system health and has been used for centuries as a flavorful spice in cooking. In developing countries, a large portion of the population relies on phytomedicine due to limited access to conventional pharmaceuticals, as herbal remedies are often more affordable and, in some cases, more effective for treating specific ailments (Singh and Gohil, 2024). Among the vast array of medicinal plants, black cumin is distinguished as one of the most highly valued and nutrient-rich herbs in human history. Among various compounds of black cumin, thymoquinone is very important and has been identified as the main component with anti-inflammatory, anti-cancer, and antioxidant properties (Sarkar et al., 2021; Sadeghi et al., 2023). Historical and folk practices have utilized black cumin seeds as a remedy for an array of health conditions, including hypertension, diabetes, and gynecological disorders. The seed's ability to address these ailments has

earned black cumin a distinguished place in the world of natural healing. In addition to its medicinal importance, the seed yield of black cumin is a critical trait for agricultural and pharmaceutical pursuits. Various genotypes of black cumin are distributed across diverse geographic regions, particularly within the Mediterranean basin and western Asia, reflecting its remarkable adaptability and genetic diversity (Fikre et al., 2023). Black cumin has gained increasing recognition in both the food and pharmaceutical industries, thanks to its wide-ranging applications. It is a plant of immense interest for pharmaceutical and functional food developers, owing to the bioactive phytochemicals it contains (Alu'datt et al., 2024). These compounds exhibit a variety of pharmacological properties, making black cumin a cornerstone for researchers working on the development of medicinal plant-based products.

Black cumin is widely distributed across various regions of Iran, thriving at diverse altitudes. This broad geographical range reflects significant genetic diversity within black cumin populations, highlighting the importance of systematic conservation efforts. Mehri et al. (2022) utilized 24 ISSR molecular markers to assess the genetic diversity among 28 Iranian black cumin genotypes, revealing significant polymorphism and classifying the genotypes into three main groups. This study provides valuable insights for breeding programs aiming to enhance desirable traits. A study by Faravani et al. (2013) investigated the variation in agronomic and anatomic characteristics of *Nigella sativa* landraces

in Khorasan, Iran. The research highlighted significant differences in traits such as plant height, number of branches, and seed yield, emphasizing the potential for selecting superior genotypes for cultivation. Research conducted by Neghab and Panahi, (2017) involved the molecular characterization of Iranian black cumin accessions using RAPD markers. The findings demonstrated high genetic diversity among the genotypes, which is crucial for germplasm conservation and future breeding efforts. Despite its significance, research on the genetic diversity of black cumin populations remains limited, creating a gap in understanding this valuable species. Insufficient knowledge about its genetic variation and the relationships between different populations hampers the widespread cultivation and development of improved cultivars. Genetic diversity is crucial for identifying superior genotypes, improving breeding programs, and ensuring sustainable cultivation. Understanding this diversity is key to producing high-performing varieties and meeting the growing demand for medicinal plants in the pharmaceutical and nutraceutical industries, while also preserving the species for future generations (Singer et al., 2021; Hafeez et al., 2023).

Studies have demonstrated significant genetic variation in key traits of black cumin, including biomass, capsule seed count, yield performance, and the weight of a thousand seeds (Gashaw et al., 2020; Shesuleiman et al., 2021). These findings suggest that by evaluating such agronomic traits, especially those directly linked to seed yield, plant breeders can develop focused strategies to

enhance black cumin varieties. . Despite the recognized importance of medicinal plants like black cumin, efforts to implement comprehensive breeding programs for this species have been limited. Moreover, the existing studies on the genetic diversity of black cumin are few and far from exhaustive. The identification and characterization of superior genotypes remain largely underexplored, highlighting a critical gap in the systematic utilization of its genetic resources (Panahi et al., 2024). To address this gap, the analysis of untapped genetic diversity within black cumin populations is required to enhance crop improvement. Despite favorable conditions for cultivating medicinal plants, land allocated for their production remains limited, highlighting the need for plant breeding to expand their reach. While breeding for medicinal plants has traditionally lagged behind major crops, breeders are now tapping into genetic diversity to improve yield and sustainability (Dsouza et al., 2025). Research on black cumin has revealed significant genetic variation in traits like seed yield and biomass, offering opportunities to improve varieties. However, comprehensive breeding programs for black cumin are still limited, and the identification of superior genotypes remains underexplored. To address this gap, further analysis of genetic diversity in black cumin populations, particularly from diverse regions of Iran, is essential for advancing breeding efforts. Breeding black cumin, a valuable medicinal plant, faces specific challenges that set it apart from major crop species. One of the key challenges is the potential for chemical changes or fluctuations

in the levels of medicinal compounds during the breeding process. Since the therapeutic properties of black cumin are largely attributed to compounds like thymoquinone, any genetic modifications that impact these compounds could compromise its medicinal value. Consequently, it is vital to observe and guarantee that these important compounds are preserved or improved when choosing high-quality genotypes for breeding. This consideration is vital for the success of breeding programs aimed at improving black cumin, as the quality and efficacy of the medicinal compounds must be preserved while also enhancing traits like yield and disease resistance. Furthermore, research on genetic diversity and the identification of superior genotypes, particularly those with medicinal attributes, represent a relatively neglected field that necessitates increased focus to fully realize the potential of black cumin as both an agricultural product and a therapeutic resource. Focusing on the economic applications of black cumin is crucial, particularly in specialized fields like producing black seed oil and medicinal compounds. A targeted approach, such as comparative studies of different genotypes, can help identify those with superior characteristics for high-quality oil production or enhanced medicinal properties, such as higher concentrations of bioactive compounds like thymoquinone. These investigations can directly contribute to practical developments in the industry, improving the efficiency and quality of black seed-based products. By selecting genotypes optimized for specific economic uses, breeding programs can drive advancements

in the pharmaceutical and nutraceutical sectors, meeting the growing demand for effective, natural remedies. Researching the morphological variation of black cumin enhances understanding of its genetic diversity, aiding taxonomy and conservation efforts in botany. It also provides critical insights for breeding programs, enabling the selection of traits for improved yield, disease resistance, and adaptability. Current research aims to investigate the genetic variation of black cumin genotypes originated from diverse regions of Iran, using a detailed examination of morphological traits.

## **Material and methods**

### *Trial and traits*

A total of twenty-seven accessions of black cumin genotypes (Table 1), were collected from various regions across Iran to assess their genetic diversity. The germplasms were cultivated under controlled greenhouse (16/8 hours for day/night duration, 70 % humidity, and 25/20 °C for day/night temperature) conditions using a randomized complete block design (with four replicates to ensure the reliability and accuracy of the results. Each experimental unit consisted of 0.25-meter diameter pots filled with soil that had been prepared and enriched with farmyard manure to provide the necessary nutrients for optimal plant growth. To account for potential variability in germination and early growth, ten seeds from each genotype were initially sown per pot. Once the seedlings had established and reached an initial stage of growth, the number of plants in each pot was reduced to five. This thinning process was implemented

to optimize growth conditions, minimize competition among plants, and maintain uniformity across experimental units. Throughout the growing period, standard agronomic practices were meticulously followed to ensure favorable conditions for plant development. Manual surface irrigation was conducted every two days to maintain consistent soil moisture levels for maintaining field capacity between 60-80%, while weeds were carefully removed by hand to prevent interference with plant growth. These measures helped create a controlled environment that allowed for the accurate assessment of plant traits. Data were recorded for a range of agronomic and morphological traits according to Moradzadeh et al. (2021), across the replications to comprehensively evaluate the genetic diversity of the collected accessions. The traits measured included plant height (PH), length of flowering stem (LFS), stem diameter (SD), internodes of the main stem (IMS), internodes to the first flower (IFF), leaf area (LA), number of seeds per follicle (NSF), number of follicles per plant (NFP), capsule weight (CW), and dry weight of shoot (DWS). Additional measurements included the length of internodes (LI), leaf width (LW), leaf length (LL), chlorophyll content (CC), follicle width (FW), follicle length (FL), thousand-seed weight (TSW), seed yield (SY), seed length (SL), and seed width (SW).

#### *Data analysis*

The genetic diversity of black cumin genotypes was assessed through multivariate statistical analysis, with a tree diagram constructed to visualize the relationships and

clustering patterns among the genotypes. Agronomic performance was analyzed for each cluster to identify the contribution of various traits to the observed genetic diversity. To evaluate the distribution of measured traits across the black cumin genotypes, the Kolmogorov–Smirnov procedure was used via the Normality statement in MINITAB software version 17.0 (Minitab Inc., USA). This test ensured the normality of the data, a critical assumption for subsequent statistical analyses. Agglomerative hierarchical clustering was done to categorize the black cumin genotypes and measured traits according to standardized squared Euclidean distances derived from the dataset. This method is the most common type of hierarchical clustering used to group individuals in clusters based on their similarity, and was advised by many researchers for evaluation of genetic variations in plants (Sabaghnia et al., 2012 and 2015). The squared Euclidean distances were computed and subsequently merged using Ward’s minimum variance method, which optimizes the clustering by minimizing within-cluster variance. The resulting dendrograms visually represented the patterns of similarity and relationships among genotypes and traits. These analyses were carried out using the Mult/Exploratory statement in STATISTICA software version 10.0 (TIBCO Inc., USA). To determine the optimal number of clusters and evaluate significant partitions within the dendrograms, multi-variate ANOVA was applied to the original dataset. Various statistical measures, including statistics of Hotelling, Wilks, Pillai, and Roy, were

**Table 1.** Identification and certain geographic characteristics of the collected black cumin genotypes

No.	Origin	Province	Altitude (m)	Longitude/ Latitude
G1	Ardabil-I	Ardabil	1377	38°15'N 48°17'E
G2	Ardabil-II	Ardabil	1377	38°15'N 48°17'E
G3	Amlash	Gilan	15	37°05'N 50°11'E
G4	Ahwaz	Khuzestan	12	31°18'N 48°40'E
G5	Arak	Markazi	1700	34°05'N 49°41'E
G6	Isfahan-I	Isfahan	1570	32°39'N 51°40'E
G7	Isfahan-II	Isfahan	1570	32°39'N 51°40'E
G8	Borujen	Chaharmahal Bakhtiari	2200	31°58'N 51°17'E
G9	Birjand	South Khorasan	1491	33°05'N 59°10'E
G10	Piranshahr	East Azarbaijan	1460	36°41'N 45°08'E
G11	TorbathHeydariye	Razavi Khorasan	1333	35°17'N 59°13'E
G12	Takestan	Zanjan	1265	36°04'N 49°41'E
G13	Semirom	Isfahan	2400	31°24'N 51°34'E
G14	Sarbisheh	South Khorasan	1820	32°34'N 59°47'E
G15	Shiraz	Fras	1514	29°36'N 52°32'E
G16	Qazvin	Qazvin	1316	36°17'N 50°00'E
G17	Karaj	Alborz	1300	35°49'N 50°56'E
G18	Lordegan	Lorestan	1700	31°30'N 50°50'E
G19	Lorestan	Lorestan	2300	32°28'N 46°49'E
G20	Kashan	Isfahan	945	33°58'N 51°25'E
G21	Kashmar	Razavi Khorasan	1215	35°14'N 58°27'E
G22	Kordestan	Kordestan	1500	35°19'N 47°22'E
G23	Mashhad-I	Razavi Khorasan	1050	36°19'N 59°32'E
G24	Mashhad-II	Razavi Khorasan	1050	36°19'N 59°32'E
G25	Marivan	Kordestan	1320	35°31'N 46°10'E
G26	Miandoab	East Azarbaijan	1314	36°57'N 46°06'E
G27	Neyshabur	Razavi Khorasan	1193	36°12'N 58°47'E

computed to evaluate the meaningfulness of clustering and identify meaningful groupings. These analyses were performed using the Multivariate statement in SPSS software version 26.0 (SPSS Inc., USA). The significance thresholds determined by these tests were used as the final cutoff points for defining clusters.

### Results and discussion

Descriptive statistical indices, including the coefficient of variation (CV), are presented in Table 2. The CV values showed relatively high variabilities among the black cumin traits. High CV values ( $>20\%$ ) were observed for plant height (PH), length of flowering stem (LFS), stem diameter (SD), internodes of the main stem (IMS), internodes to the first flower (IFF), leaf area (LA), number of seeds per follicle (NSF), number of follicles per plant (NFP), capsule weight (CW), and dry weight of shoot (DWS). These high CVs indicate substantial genetic diversity in these traits, which could serve as valuable targets for future breeding programs. Moderate CV values ( $10\% < CV \leq 20\%$ ) were recorded for traits such as length of internodes (LI), leaf width (LW), leaf length (LL), chlorophyll content (CC), follicle width (FW), follicle length (FL), thousand-seed weight (TSW), and seed yield (SY). In contrast, low CV values ( $< 10\%$ ) were observed for seed length (SL) and seed width (SW), suggesting relatively lower variability for these traits among the genotypes (Table 2). The high genetic variation observed across most traits highlights the potential of black cumin germplasm for selecting and combining favorable traits to

develop improved varieties.

Supporting this finding, Mehri et al. (2022) examined various black cumin landraces and reported high CV values for yield components such as the seeds and follicles numbers, whereas relatively low CV values were seen for thousand-seed weight and seed yield. Similarly, Ahmadi et al. (2024) evaluated genetic variation among twenty black cumin genotypes and reported high magnitudes of CV for the branches and capsules of the plant, as well as plant height and yield performance. Moderate CV values were noted for the seeds of capsule and thousand-seed weight in their study. In contrast to previous findings that identified low variability in seed size (length and width), this study found high or moderate CV values for nearly all measured traits, including seed yield and yield components, so this may be due to some factors such as differences in research methods, type of genotypes, or various environmental conditions, which influences the performance of black cumin. These results emphasize the extensive genetic variability within the current germplasm, making it a promising resource for use in breeding programs aimed at enhancing agronomic performance and productivity in black cumin. This study reached different results from previous research, particularly regarding the observed genetic variation in seed size and yield components, likely due to several factors. First, differences in research methods could explain the variation, as different techniques for measuring traits (e.g., statistical approaches, marker usage) might yield varying results. Second, the selection of genotypes may have played a role; if this

**Table 2.** Descriptive statistics of the measured traits of black cumin genotypes

	Mean	SD	Range	CV
PH	28.4	11.2	37.7	39.5
LI	2.6	0.4	1.5	15.8
LFS	4.9	1.1	5.3	23.0
SD	1.5	0.6	2.3	40.4
IMS	8.5	2.7	9.4	31.8
IFF	9.5	2.6	9.7	27.6
LW	20.0	3.4	10.5	16.8
LL	40.6	6.4	23.1	15.9
LA	494	118	504	23.8
CC	1.7	0.2	1.1	12.7
FW	9.3	1.1	5.6	11.9
FL	10.0	1.4	4.8	13.5
NSF	10.7	3.6	13.0	33.4
SL	2.5	0.1	0.7	5.7
SW	1.5	0.1	0.3	4.1
NFP	6.6	4.8	16.0	73.9
CW	0.3	0.1	0.4	43.3
DWS	1.7	1.9	5.9	113.1
TSW	2.5	0.3	1.1	12.0
SY	100	12.2	49.2	12.1

SD, standard deviation; CV, coefficient of variation

study included different or more diverse genotypes than previous studies, it would naturally lead to higher observed variation. Lastly, environmental conditions could be a significant factor, as differences in soil, climate, and cultivation practices can impact plant traits and overall performance. These factors likely influenced the high genetic variation found in this study, highlighting the potential of the germplasm for breeding

programs aimed at improving black cumin. A dendrogram was constructed to identify patterns and relationships among black cumin genotypes (Fig. 1).

The optimal cutoff point for clustering was determined using Wilks, Hotelling, Pillai, and Roy statistics (Table 3), all of which were statistically significant, confirming the robustness of the clustering approach.

Various cutoff points, ranging from two to 23



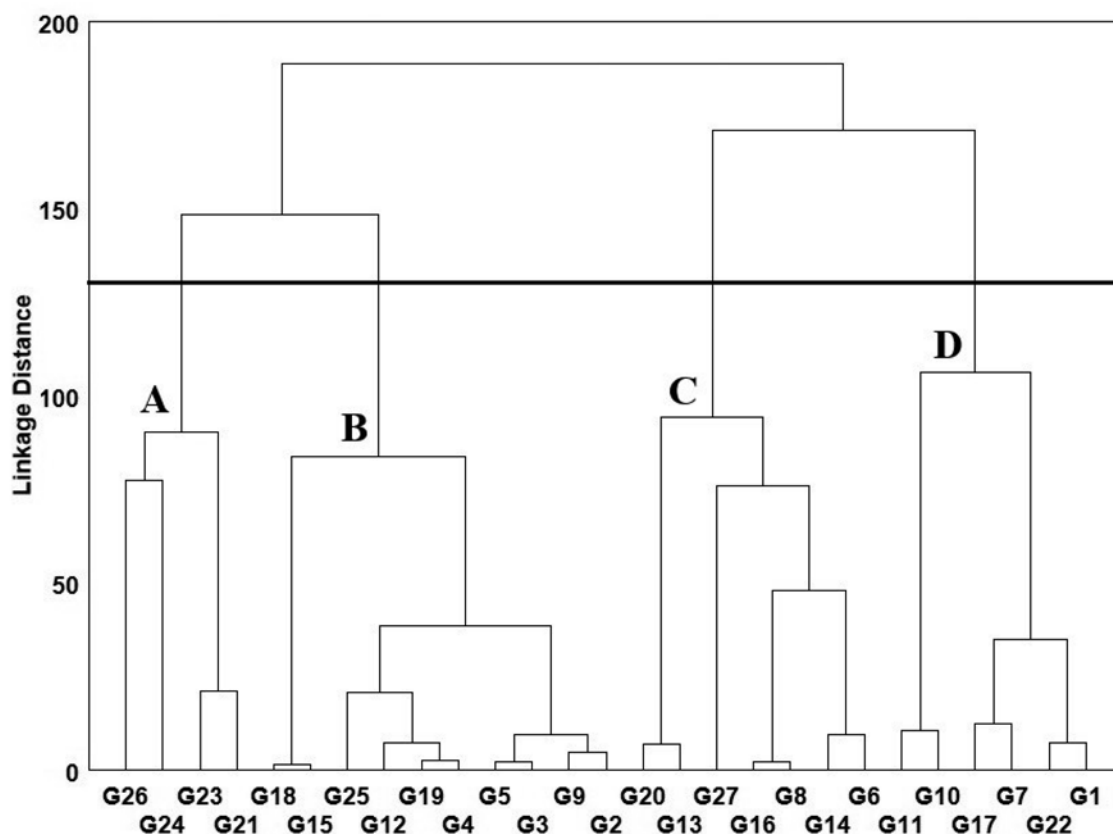


Fig. 1. Dendrogram of cluster analysis for 27 genotypes of black cumin

**Table 3.** Multivariate indices to determine the cutoff point for classification of genotypes

Statistics	Value	F†	Hypothesis df‡	Error df	Sig.
Pillai's Trace	2.66	2.37	60	18	0.02
Wilks' Lambda	0.02	39.27	30	2	0.01
Hotelling's Trace	93.23	2.95	60	8	0.03
Roy's Largest Root	33.25	9.97	20	6	0.00

†F, statistic of F ratio; ‡df, degrees of freedom

clusters, were tested, with genotypes within each cluster used as replicates for multivariate analysis across all traits. Based on this analysis, the dendrogram divided the black cumin genotypes into four distinct clusters. The average traits of the identified clusters are summarized in Table 4, providing a detailed characterization of each cluster. Cluster A includes four genotypes (G21,

G23, G24, and G26) from Razavi Khorasan (Kashmar, Mashhad-I, and Mashhad-II) and East Azarbaijan (Miandoab). This cluster is notable for its superior seed yield and high values across most traits, though it exhibits relatively low values for seed width and thousand-seed weight. These genotypes show strong potential for yield improvement, making them prime candidates for

**Table 4.** The averages of traits for black cumin genotypes in four identified clusters

	A	B	C	D
PH	45.03	28.30	25.60	20.98
LI	3.23	2.58	2.48	2.21
LFS	6.27	4.80	4.78	4.18
SD	2.44	1.45	1.50	1.18
IMS	12.17	8.64	7.68	6.95
IFF	12.63	9.71	8.72	8.05
LW	23.93	21.76	18.14	16.74
LL	49.96	42.67	38.57	33.08
LA	692.9	537.5	447.0	345.7
CC	1.84	1.74	1.73	1.64
FW	10.69	9.48	9.23	8.27
FL	11.72	9.88	9.99	9.24
NSF	15.95	10.21	10.29	8.48
SL	2.65	2.56	2.51	2.44
SW	1.48	1.52	1.51	1.47
NFP	14.20	5.56	5.86	3.91
CW	0.42	0.27	0.26	0.21
DWS	4.27	1.37	1.51	0.64
TSW	2.16	2.55	2.59	2.53
SY	107.4	96.8	107.5	93.4

breeding programs focused on maximizing productivity. Cluster B consists of ten genotypes (G2, G3, G4, G5, G9, G12, G15, G18, G19, and G25). These genotypes display low to moderate values for most traits but stand out for high values in leaf width, seed width, and thousand-seed weight. While their overall yield potential is lower, their larger seed size and higher thousand-seed weight could make them valuable in markets prioritizing these traits. Cluster

C includes seven genotypes (G6, G8, G13, G14, G16, G20, and G27), which demonstrate high seed yield performance. These genotypes also show high values for leaf width and thousand-seed weight, along with moderate values for follicle width and chlorophyll content. The combination of these traits suggests that the high leaf width and thousand-seed weight may compensate for the moderate follicle width and chlorophyll content. This cluster represents a balanced

group with promising potential for breeding programs targeting both high yield and marketable traits. Cluster D consists of six genotypes (G1, G7, G10, G11, G17, and G22), which are characterized by low yield performance but high thousand-seed weight, the highest among all clusters. These genotypes are particularly notable for their large seed size, making them appealing for markets that prioritize seed size and weight. Despite their low yield, these genotypes could serve as valuable resources for breeding programs aimed at enhancing seed size for specific market demands.

The clustering analysis reveals clear genetic diversity within the black cumin germplasm, with each cluster exhibiting distinct agronomic and morphological traits (Golkar and Nourbakhsh, 2019). Genotypes in Cluster A and Cluster C stand out for their high yield potential, while those in Cluster B and Cluster D are valued for marketable traits such as larger seed size and higher thousand-seed weight. This information can inform breeding strategies aimed at developing high-yielding, resilient, and market-specific varieties of black cumin. The results of Golkar and Nourbakhsh (2019) regarding genetic diversity and agronomic traits in *Nigella sativa* are consistent with trends observed in other recent studies of medicinal plants. Genetic variation in agronomic traits, bioactive compound content, and the use of molecular markers are common themes in plant breeding research across different medicinal species. This study's findings contribute to the broader context of plant breeding by emphasizing the importance of both agronomic performance and medicinal properties in

the selection of superior genotypes. This approach supports the advancement of breeding programs aimed at achieving crop yields and pharmaceutical applications.

A dendrogram was generated to analyze the structural relationships among black cumin traits (Fig. 2).

The cutoff point for clustering was validated using four multivariate analysis of variance indices, Wilks, Hotelling, Pillai, and Roy, all of which were statistically significant and confirmed the selected cutoff point (Table 5). Based on these results, traits were grouped into five distinct categories. Group I includes seed width and thousand-seed weight, traits closely related to seed size. Group II comprises leaf-related traits such as leaf area, leaf width, and leaf length, which reflect the plant's vegetative characteristics. Group III consists of seed yield, seed length, flowering stem length, follicle width, and internode length, indicating a mix of yield-related and structural traits. Group IV focuses on chlorophyll content, a key indicator of the plant's photosynthetic capacity. Group V encompasses a variety of traits, including plant height, stem diameter, internode length, capsule weight, seeds per follicle, number of follicles, dry weight of the shoot, and follicle length. While previous studies by Kara et al. (2015) and Nurozi-Boldaji et al. (2024) emphasized traits like seeds per follicle, number of follicles, and thousand-seed weight as key contributors to black cumin performance, this study found seed yield clustered separately from these traits. Instead, seed yield was strongly associated with traits such as seed length, flowering stem length, follicle width, and internode

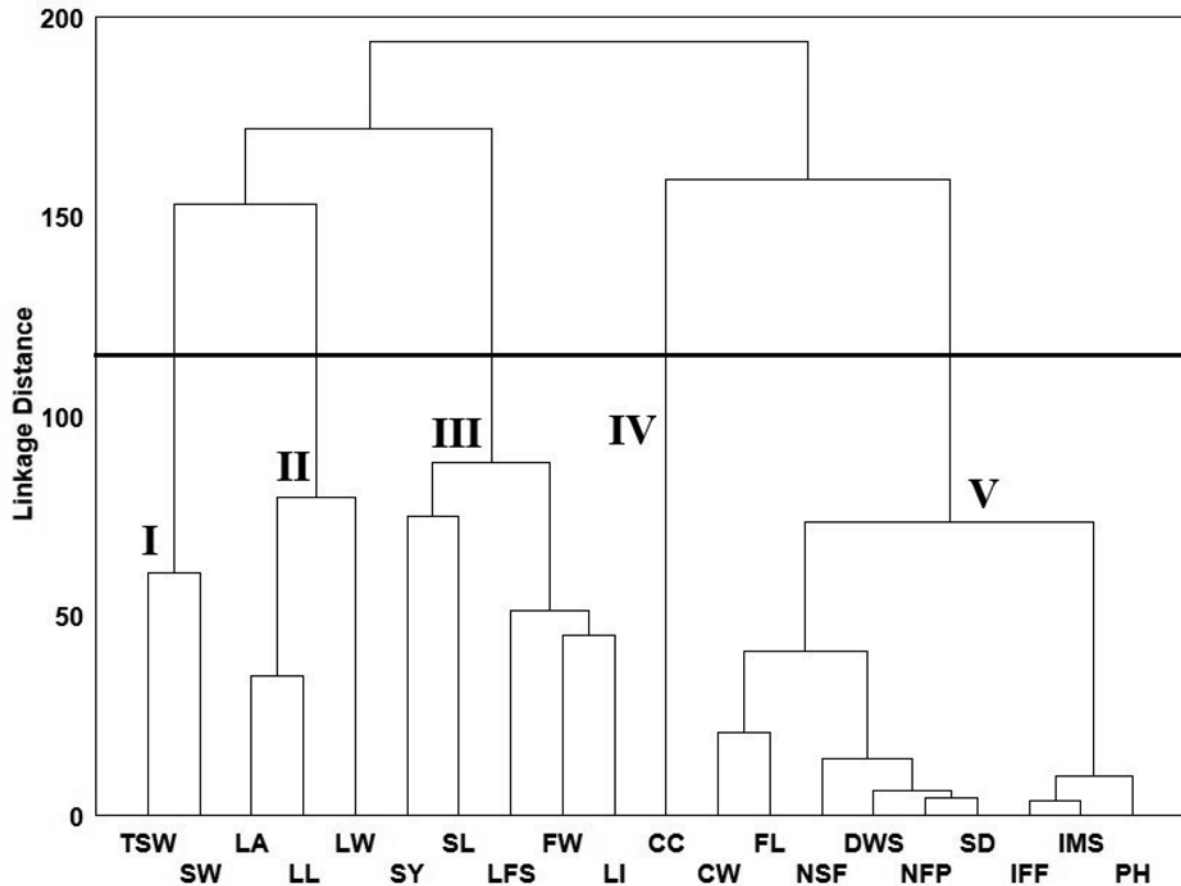


Fig. 2. Dendrogram of cluster analysis for measured traits of black cumin

**Table 5.** Multivariate indices to determine the cutoff point of dendrogram for categorizing traits

Statistics	Value	F†	Hypothesis df‡	Error df	Sig.
Pillai's Trace	12.66	4.34	60	16	0.00
Wilks' Lambda	0.02	41.22	60	6	0.00
Hotelling's Trace	3357.35	154.72	60	8	0.00
Roy's Largest Root	3573.37	852.85	15	4	0.00

†F, statistic of F ratio; ‡df, degrees of freedom

length. This suggests that traits indirectly related to yield can play a crucial role in improving black cumin productivity and should be considered in breeding programs. Ahmadi et al. (2024) found a positive relationship between seed yield and the number of branches and capsules but did not

identify significant relationships with seeds per follicle or thousand-seed weight. These discrepancies highlight the complexity of yield-related traits and suggest that black cumin's yield performance cannot be fully explained by traditional yield components alone. Finally, genetic variations among the

genotypes used in each study might also influence how traits are correlated with seed yield. Further investigation into these aspects could provide a clearer understanding of the complex relationships between traits and seed yield in black cumin.

### Conclusions

The genotypes examined in this study demonstrated a variety of valuable characteristics that can be leveraged for different breeding programs. Among these, the four genotypes from Kashmar, Mashhad-I, Mashhad-II, and Miandoab performed particularly well and are recommended for farmers due to their superior seed yield performance. These genotypes, with their desirable agronomic traits, are promising candidates for future breeding programs aimed at enhancing the yield and quality of black cumin.

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