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Genotype × trait biplot analysis of genetic diversity and trait associations in some fenugreek landraces

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Abstract

Objective: Fenugreek (*Trigonella foenum-graecum* L.), as an economically and culturally significant legume, is valued for its diverse uses in food, medicine, and sustainable agriculture. This study aimed to assess the genetic diversity among 26 fenugreek landraces collected from diverse agroecological zones in Iran.

Methods: The landraces were evaluated under field conditions using a randomized complete block design with three replicates. Morphological traits, including plant height, number of lateral branches, leaf dimensions, pods per plant, and shoot biomass, were recorded alongside physiological traits such as chlorophyll-a and chlorophyll-b content. Data analysis involved a genotype-by-trait biplot approach based on principal component analysis to visualize genotype × trait interactions and to identify traits contributing most significantly to yield and biomass.

Results: The two components explained 66% of the total phenotypic variance, revealing additive and crossover associations among landraces and traits. Key positive trait associations included shoot biomass with plant height, chlorophyll-a with chlorophyll-b, lateral leaf width with lateral leaf area, and a cluster of branching and pod number traits. Genotype-specific patterns highlighted the superiority of genotype 6 (Sarab) and genotype 8 (Tabriz-II) for the shoot biomass and middle leaf area. In contrast, genotype 9 (Tehran-II) excelled in branching and pod production. These landraces also exhibited high stability and representativeness in multivariate trait space, positioning them as promising candidates for breeding programs and commercial release.

Conclusion: This study confirms that middle leaf area, number of lateral branches, lateral leaf length, pods per plant, and plant height are critical discriminative and representative traits for fenugreek improvement. The

integration of morphological and physiological traits in a multivariate framework provides a robust basis for indirect selection and ideotype development. Furthermore, the results highlight the importance of considering genotype \times trait interactions to prevent misleading selection decisions.

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Introduction

The genus *Trigonella*, belonging to the Fabaceae family, includes several economically important species, with fenugreek (*Trigonella foenum-graecum* L.) being one of the most prominent. It is a diploid species ($2n = 16$), predominantly self-pollinated, and adapted to a wide range of climatic conditions (Valizadeh 2018). This annual plant is native to the Eastern Mediterranean region but has been widely cultivated across diverse areas of the world (Roba and Simion 2022; Azizi *et al.* 2025). Major cultivation areas include the Indian subcontinent, Iran, various African countries, and some parts of Europe (Kamalvand *et al.* 2022). In some regions of the world, fenugreek holds significant traditional value, serving multiple purposes within agricultural and culinary systems. The diverse uses of this crop include consumption of fresh leaves as vegetables, dried leaves as flavoring agents, seed utilization in spices and traditional medicines, and extraction for health products. Traditional medicinal uses of fenugreek seeds target a variety of ailments, including digestive problems, inflammation, diabetes, and cardiovascular diseases. Fenugreek is often employed as a rotation crop that enhances soil fertility and structure, contributing to sustainable farming practices (Mechri *et al.* 2025). Additionally, fenugreek provides considerable income for smallholder farmers and is incorporated into local diets; its flour enhances the flavor and texture of staple foods such as tef-injera, particularly in cooler regions where tef is a dietary mainstay (Legassa *et al.* 2022). The flour is also a key ingredient in various spice blends, and traditional preparations include a nutritious drink made by soaking fenugreek powder overnight and mixing it with honey. Furthermore, in some communities, fenugreek is used as an alternative infant food when milk is scarce (Dhull *et al.* 2021).

Fenugreek cultivation in many areas parallels that of some cool-season pulses like chickpea and lentil (Duc *et al.* 2015). Despite low ranking in production volume among highland pulses, fenugreek stands out as the leading cash-generating legume crop. This economic significance presents a valuable

opportunity to expand market access, especially for small-scale farmers. The flexible uses of crop at various growth stages, ranging from green manure and leafy vegetables to seeds, further enhance its agricultural value (Narayana *et al.* 2022).

Despite the rich genetic resources of Iran for fenugreek, traditional cultivation practices prevail, with minimal use of improved varieties. This stagnation poses a risk of losing valuable genetic diversity accumulated through centuries of local cultivation (Khoury *et al.* 2022). A critical barrier to fenugreek improvement is the limited understanding of its genetic variation. Effective breeding requires comprehensive knowledge of genetic diversity, trait associations, and heritability; data that are currently scarce for fenugreek genotypes (Berhe *et al.* 2025). Also, fenugreek productivity remains low due to the absence of suboptimal agronomic practices. Yield performance, as a complex quantitative trait, depends on multiple components, necessitating detailed analysis to identify those that contribute most significantly to performance (Sabaghnia *et al.* 2024a). Correlation studies offer initial insights into trait relationships, while path coefficient analysis is instrumental in dissecting the effects of individual traits on yield performance, thereby facilitating more precise selection in breeding programs (Molaei *et al.* 2023).

The study by Al-Maamari *et al.* (2020) assessed the morphological and genetic diversity of 20 fenugreek accessions and indicated variability in stem length, branches, leaf area, pod length, and seed weight. Also, they found high heritability for the seed number, leaf area, and leaf number, indicating potential for selection, providing valuable insights for breeding programs aimed at improving fenugreek cultivation. Meena *et al.* (2021) evaluated 17 fenugreek landraces for genetic variability and showed high heritability and genetic advance for traits such as grain yield performance and plant height. They grouped fenugreek landraces into six clusters, with grain yield per plot contributing the most to genetic divergence, so they concluded that improving grain yield in fenugreek can be achieved by selecting for traits like pod number, seed number, plant height, and primary branches. A study on the genetic variability of Ethiopian fenugreek accessions assessed 160 accessions and revealed that the first five components explained 66% of the total variation, with key contributors being seeds per pod, pods per plant, and branch numbers, while cluster analysis grouped accessions into two major clusters, indicating substantial genetic diversity and potential for crop improvement through selection or hybridization (Roba and Mohammed 2024).

Given the quantitative nature of yield traits and their sensitivity to environmental factors, it is crucial to determine the extent of their variation. Therefore, this research was undertaken to assess the variability present in the collected germplasm and to analyze the associations among traits critical for yield improvement, thus supporting effective fenugreek breeding programs. Given these

considerations, the present study aimed to (i) assess genetic variation among Iranian fenugreek landraces, (ii) examine associations between agronomic and morphological traits, and (iii) identify superior landraces for potential commercial release and future breeding efforts.

Materials and Methods

Trial

A collection of 26 fenugreek landraces from various regions throughout Iran (Table 1), were evaluated using a randomized complete block design with three replicates. After routine soil preparation, including plowing and leveling in April, experimental plots measuring 50 by 80 cm were established. Throughout the growing season, uniform irrigation was maintained in all plots, and manual weeding was regularly performed to ensure optimal growth conditions. Fertilization followed local recommendations, applying 25 kg ha⁻¹ N as urea, 60 kg ha⁻¹ P₂O₅ in the form of triple super phosphate, and 25 kg ha⁻¹ K as potassium sulfate. Continuous monitoring for pest and disease infestations was carried out, with appropriate control measures implemented as needed.

Traits

At the midpoint of flowering, seven plants per plot were randomly selected to measure the morphological and physiological characteristics. These included plant height, number of lateral branches, middle leaf width, middle leaf area, lateral leaf length, lateral leaf width, lateral leaf area, and pods per plant. Leaf sampling was taken from the mid-portion and upper third of newly extending shoots, ensuring that fully expanded leaves are selected. Middle leaves were taken from the mid-stem nodes of the main shoot, while lateral leaves were taken from lateral branches. Harvest was scheduled for August, coinciding with pod maturation, indicated by yellowing pods and dried foliage. Post-harvest assessment for measuring shoot biomass, was performed on seven randomly selected plants. Standardized measuring instruments were employed including manual counting, a stainless-steel ruler with ±1.0 mm precision, a digital scale with ±0.01 g accuracy, calipers with ±0.1 mm resolution, and a leaf area meter (AM-3000, ADC BioScientific Ltd). Photosynthetic pigments such as chlorophyll-a and chlorophyll-b were measured regarding the Arnon (1941) protocol, as adapted by Rostami *et al.* (2022).

Data analysis

Data cleaning for checking missing values and exploring outliers was done and after normality assessment of errors, symmetrical scaling and standardization of dataset was performed for traits

Table 1. Geographic coordinates of the collected fenugreek landraces.

| No. | Name | Coordinates | Elevation | No. | Name | Coordinates | Elevation |
|-----|-------------|-----------------|-----------|-----|------------|-----------------|-----------|
| 1 | Mashhad | 36°19'N 59°32'E | 995 | 14 | Kerman | 30°15'N 57°03'E | 1764 |
| 2 | Gorgan | 36°50'N 54°26'E | 155 | 15 | Kashmar | 35°14'N 58°27'E | 1063 |
| 3 | Bushehr | 28°55'N 50°51'E | 18 | 16 | Mughan-I | 39°38'N 47°54'E | 33 |
| 4 | Ardestan | 33°22'N 52°22'E | 1207 | 17 | Jahrom | 28°30'N 53°34'E | 1050 |
| 5 | Rezvanshahr | 37°32'N 49°08'E | 5 | 18 | Mughan-II | 39°38'N 47°54'E | 33 |
| 6 | Sarab | 37°56'N 47°32'E | 1650 | 19 | Ardabil | 38°15'N 48°17'E | 1351 |
| 7 | Meshgin-I | 38°23'N 47°40'E | 1400 | 20 | Urmia-II | 37°32'N 45°03'E | 1332 |
| 8 | Tabriz-II | 38°04'N 46°17'E | 1351 | 21 | Tabriz-III | 38°04'N 46°17'E | 1351 |
| 9 | Tehran-II | 35°41'N 51°23'E | 900 | 22 | Tehran-I | 35°41'N 51°23'E | 900 |
| 10 | Urmia-I | 37°32'N 45°03'E | 1332 | 23 | Rafsanjan | 30°23'N 55°59'E | 1527 |
| 11 | Isfahan | 32°39'N 51°40'E | 1574 | 24 | Meshgin-II | 38°23'N 47°40'E | 1400 |
| 12 | Khansar | 33°13'N 50°18'E | 2215 | 25 | Khalkhal | 37°36'N 48°31'E | 2243 |
| 13 | Tabriz-I | 38°04'N 46°17'E | 1351 | 26 | Kiashahr | 37°25'N 49°56'E | 27 |

which had various scales. Normality of distributions for measured traits across the landraces was assessed using the Shapiro-Wilk test implemented in Minitab 18.0 application (Minitab Inc., USA). Data were analyzed using the genotype-by-trait biplot approach (Yan 2024) in the GGEbiplot software to explore the interaction structure between landraces (entries) and traits (testers). Thus, the Scaling = 1, Transform = 0, and Centering = 2 options were applied. The model was expressed as:

$$\frac{Y_{ij} - \bar{Y}_j}{\delta_j} = \sum_{n=1}^2 \Phi_n \Psi_{in} \Omega_{jn} + \varepsilon_{ij}$$

Where, Y_{ij} is the average of genotype i for trait j , \bar{Y}_j is the mean of trait j , δ_j is the variability as SD of each trait, Φ_n is the eigenvalue for component n in the principal component analysis (PCA), Ψ_{in} and Ω_{jn} are the related amounts for genotype i and trait j on PCA n , ε_{ij} is the residual. Also, for calculating symmetrical values, the eigenvalue was adjusted via absorption of vectors (Morovati *et al.* 2024). This adjustment produced symmetric scaling, allowing both landraces and traits to be displayed proportionally in the biplot. This approach enabled a visual exploration of the relationships among landraces, traits, and their interactive effects. Each genotype and trait were represented by a specific symbol, facilitating graphical interpretation of genotype × trait relationships and their interactions.

Results

According to the descriptive statistics for the fenugreek traits (Table 2), plant height and shoot height showed moderate variation, with means of 52.5 cm and 43.6 cm, respectively, while the number of

Table 2. Descriptive statistics of the fenugreek traits in this study.

| Trait | Median | Mean | CV | Min. | Max. | SD |
|-------|--------|-------|-------|-------|--------|-------|
| PH | 53.09 | 52.49 | 13.68 | 32.76 | 65.90 | 7.18 |
| SH | 43.78 | 43.59 | 16.20 | 22.46 | 56.71 | 7.06 |
| RL | 9.07 | 8.80 | 11.54 | 6.42 | 10.57 | 1.02 |
| LB | 1.22 | 1.18 | 54.66 | 0.37 | 2.62 | 0.65 |
| MLH | 21.94 | 22.12 | 7.26 | 19.73 | 25.96 | 1.61 |
| MLW | 12.36 | 12.55 | 9.92 | 10.02 | 15.79 | 1.24 |
| MLA | 175.0 | 192.1 | 19.29 | 143.3 | 277.5 | 37.1 |
| LLH | 23.33 | 23.53 | 5.39 | 20.71 | 25.93 | 1.27 |
| LLW | 16.03 | 16.46 | 14.84 | 11.86 | 24.22 | 2.44 |
| LLA | 256.8 | 263.1 | 15.87 | 180.0 | 369.5 | 41.8 |
| PP | 72.67 | 74.46 | 15.88 | 56.21 | 105.91 | 11.81 |
| TSW | 14.59 | 14.29 | 14.57 | 9.44 | 18.33 | 2.08 |
| Chl.a | 5.36 | 5.13 | 29.01 | 1.57 | 7.07 | 1.49 |
| Chl.b | 1.041 | 1.065 | 34.44 | 0.309 | 1.708 | 0.367 |
| CAR | 0.990 | 0.967 | 29.54 | 0.278 | 1.381 | 0.286 |

PH: Plant height, LB: Number of lateral branches, MLW: Middle leaf width, MLA: Middle leaf area, LLH: Lateral leaf length, LLW: Lateral leaf width, LLA: Lateral leaf area, PP: Pods per plant, SB: Shoot biomass, Chl-a: Chlorophyll-a, Chl-b: Chlorophyll-b.

lateral branches exhibited the highest variability (CV = 54.7%), reflecting substantial differences among landraces. Leaf traits, including middle leaf width, middle leaf area, lateral leaf length, lateral leaf width, and lateral leaf area, also varied notably, with middle leaf area and lateral leaf area showing the largest ranges and standard deviations, highlighting their potential as discriminative traits in selection. Pods per plant and shoot biomass demonstrated moderate variation among the landraces, indicating differential yield potential. Photosynthetic pigments, including chlorophyll-a, chlorophyll-b, and carotenoids, exhibited higher variability, suggesting genotypic differences in leaf pigment composition that may influence photosynthetic efficiency. These results indicate considerable phenotypic diversity among the studied fenugreek landraces, particularly for leaf morphology, reproductive traits, and pigment content, providing a good basis for further biplot analysis.

The first two principal components (PCs) from the genotype-by-trait biplot explained 66% of the variation among fenugreek landraces (Figure 1), with PC1 accounting for 47% and PC2 for 19%. This level of variation reflects both additive and crossover interaction patterns, meaning that genotype rankings differed across traits. Such crossover effects highlight the importance of considering genotype \times trait interactions when making selection decisions, consistent with previous studies in

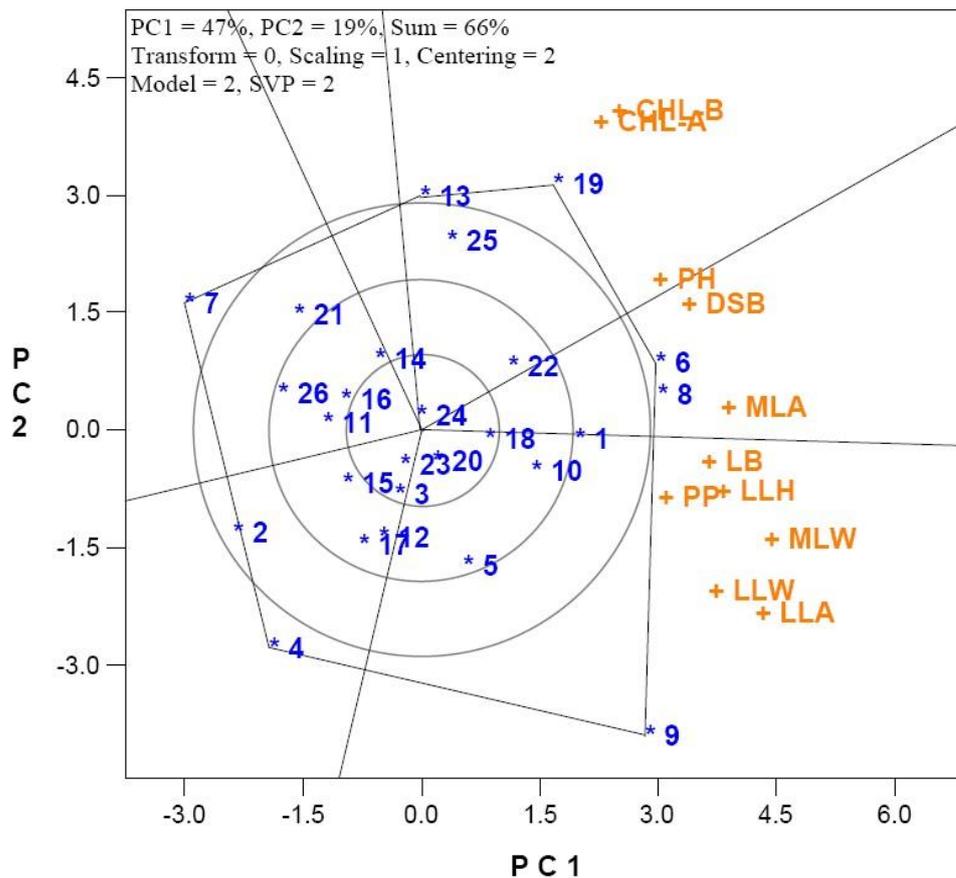


Figure 1. Which genotype of fenugreek wins which trait?

PH: Plant height, LB: Number of lateral branches, MLW: Middle leaf width, MLA: Middle leaf area, LLH: Lateral leaf length, LLW: Lateral leaf width, LLA: Lateral leaf area, PP: Pods per plant, SB: Shoot biomass, Chl-a: Chlorophyll-a, Chl-b: Chlorophyll-b.

spinach (Sabaghnia *et al.* 2016) and black cumin (Mohebodini *et al.* 2024). The biplot approach proved effective for visualizing both genotypic performance and trait relationships (Yan *et al.* 2019). In Figure 1, most traits, including number of lateral branches, middle leaf width, lateral leaf length, lateral leaf width, lateral leaf area, and pods per plant, were grouped in the sector containing landrace 9 (Tehran-II), indicating its superiority for these traits. The shoot biomass and middle leaf area formed a separate sector dominated by the landrace 6 (Sarab), followed by the landrace 8 (Tabriz-II). The landrace 19 (Ardabil) ranked highest for plant height, chlorophyll-a, and chlorophyll-b. Other vertex landraces, including 4 (Ardestan), 7 (Meshgin-I), and 13 (Tabriz-I), did not show superiority in any measured traits. These results suggest that yield performance in fenugreek is closely linked to middle leaf area. While Gutema *et al.* (2021) reported a positive effect of leaf area on biomass, Abdelhameed *et al.* (2021) highlighted the role of plant height and number of branches, reflecting some variability in trait–yield associations. Overall, the entry-by-trait biplot efficiently identified trait-specific

strengths and superior landraces. Based on these findings, the landrace 6 (Sarab, northwest Iran), followed by the landrace 8 (Tabriz-II, northwest Iran), appear most promising for commercial release, provided multi-environment trials confirm their adaptability and yield stability.

The biplot also provided clear insights into trait relationships (Figure 2). Traits pointing in the same direction are positively related, those at right angles are largely unrelated, and traits pointing in opposite directions are negatively related. Longer vectors indicate traits that contributed more strongly to differences among landraces. Strong positive associations were observed for: (i) shoot biomass with plant height; (ii) chlorophyll-a with chlorophyll-b; (iii) lateral leaf width with lateral leaf area, and (iv) number of lateral branches with middle leaf width, lateral leaf length, and pods per plant. Chlorophyll-a and chlorophyll-b were largely unrelated to number of lateral branches, middle leaf width, lateral leaf length, and pods per plant, and no negative correlations were detected. These observations were consistent with prior reports on fenugreek trait associations (Jaborova *et al.* 2021;

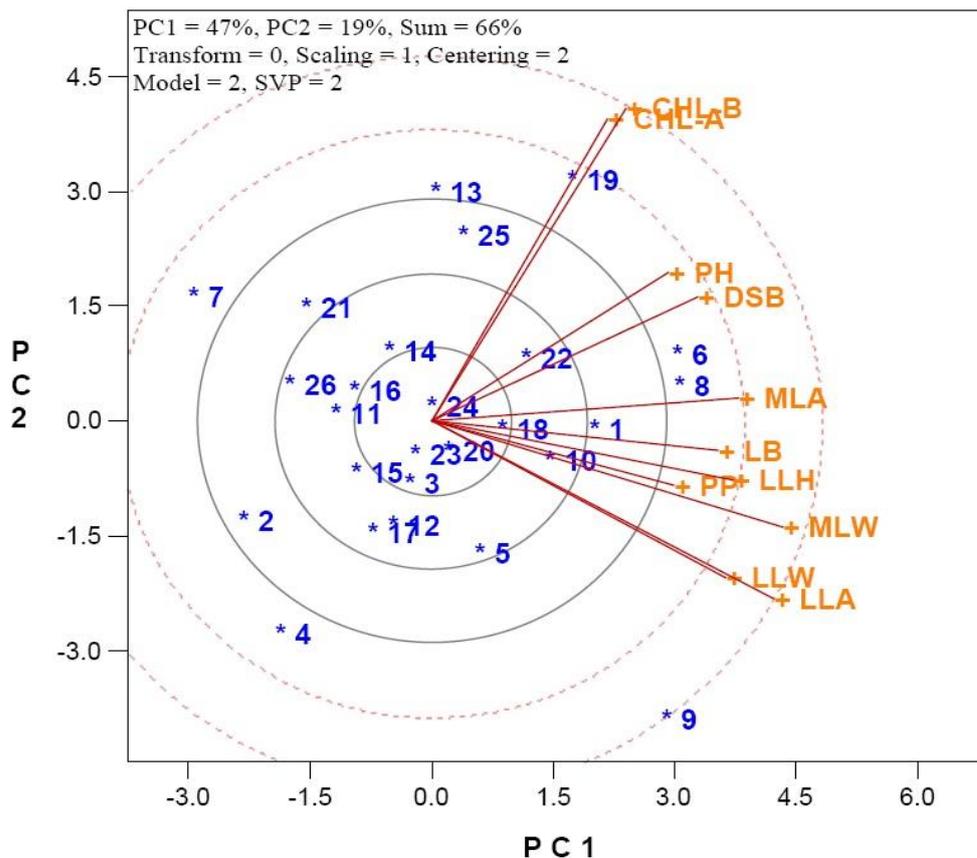


Figure 2. Associations of fenugreek traits based on genotype by trait biplot model. PH: Plant height, LB: Number of lateral branches, MLW: Middle leaf width, MLA: Middle leaf area, LLH: Lateral leaf length, LLW: Lateral leaf width, LLA: Lateral leaf area, PP: Pods per plant, SB: Shoot biomass, Chl-a: Chlorophyll-a, Chl-b: Chlorophyll-b.

Ma *et al.* 2025). While the biplot shows overall multivariate patterns, direct pairwise correlations may sometimes differ from these visual interpretations. However, pairwise correlations may be unreliable due to the indirect effects of other traits on the paired traits.

The concept of ideal landraces, those combining high performance with strong representativeness, was assessed using Figure 3. Landraces 6 and 8, followed by 1, 10, 19, and 22, were closest to the ideal point, showing high distinction ability and typical potential. Landraces 2, 4, and 7 were farthest from the ideal, marking them as least desirable. These findings can guide ideotype selection, though correlations between grain yield and other quality traits are often weak or negative (Debnath *et al.* 2024), highlighting the need for multivariate, graphical selection tools in fenugreek breeding programs.

Trait discriminative potential reflects its ability to differentiate among landraces in the biplot, with greater variation indicating higher discrimination. In the biplot (Figure 4), traits closer to the ideal trait point are considered superior, while those farther away are less desirable. Middle leaf area showed the highest discriminative potential, followed by the number of lateral branches, lateral leaf

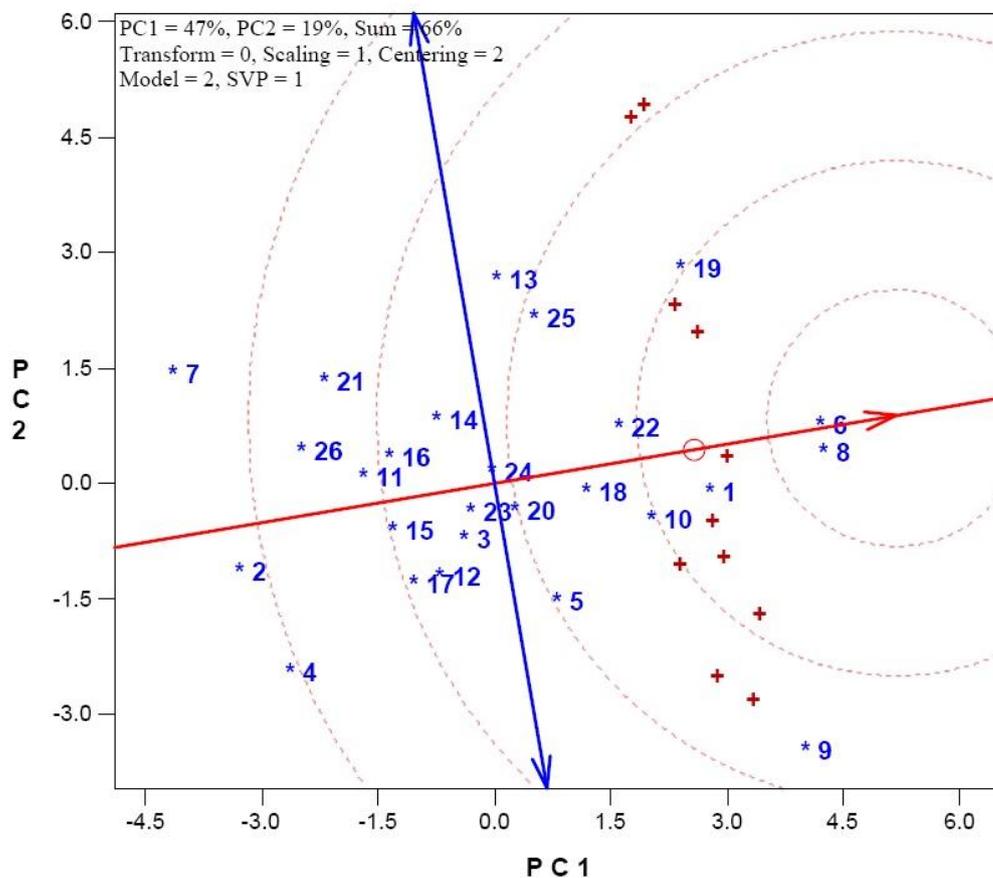


Figure 3. Ranking of fenugreek landraces based on traits.

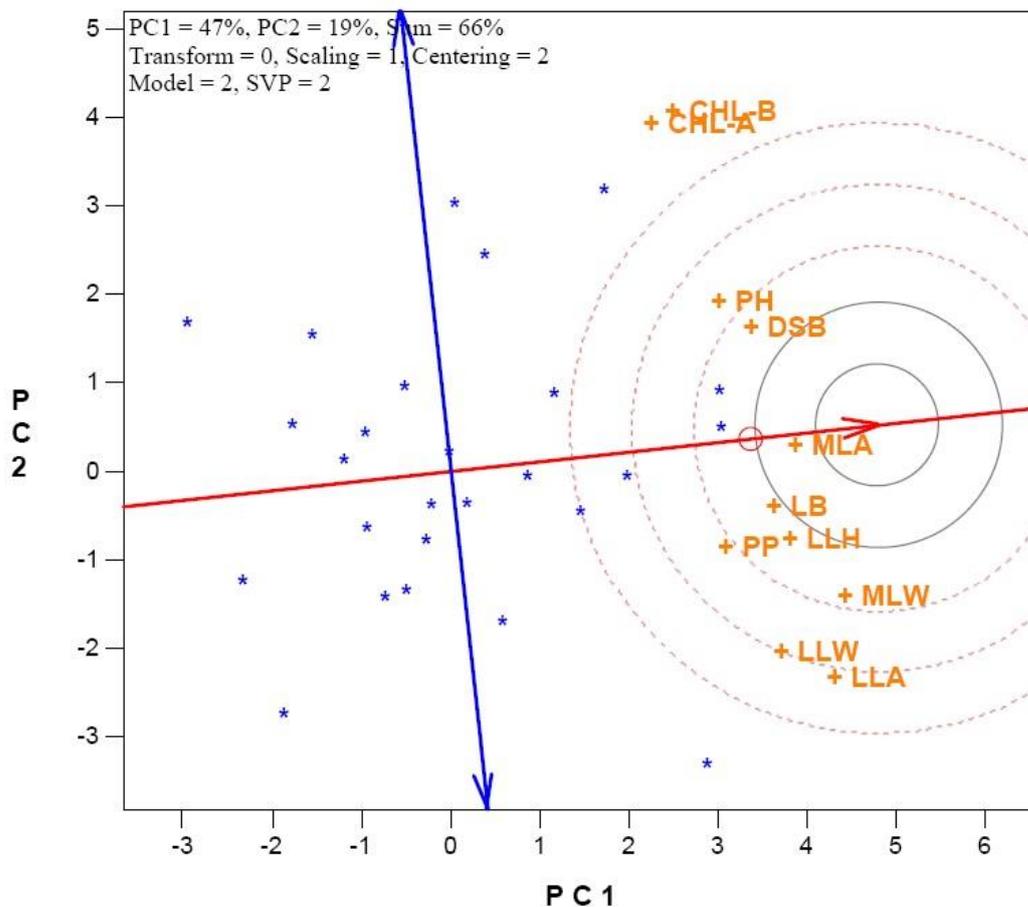


Figure 4. Ranking of fenugreek traits based on discriminative and representativeness capability. PH: Plant height, LB: Number of lateral branches, MLW: Middle leaf width, MLA: Middle leaf area, LLH: Lateral leaf length, LLW: Lateral leaf width, LLA: Lateral leaf area, PP: Pods per plant, SB: Shoot biomass, Chl-a: Chlorophyll-a, Chl-b: Chlorophyll-b.

length, pods per plant, and plant height. All traits had above-average discriminative potential, confirming their usefulness in distinguishing among landraces. Representativeness, measured by the angle with the average trait axis, was highest for middle leaf area, number of lateral branches, lateral leaf length, lateral leaf length, and plant height, meaning these traits not only discriminate well but also reflect typical patterns in the population. chlorophyll-a, chlorophyll-b, lateral leaf width, and lateral leaf area had larger angles, indicating lower representativeness. Fenugreek performance for shoot biomass is shown in Figure 5. The horizontal axis represents shoot biomass, with higher values indicating better performance. Landraces 6, 8, and 19, followed by 1, 22, 25, and 13, showed the highest shoot biomass. Landraces 4, 2, and 7 ranked lowest. The perpendicular distance from the axis reflects variability: smaller distances indicate greater stability. Landraces 6 and 8 combined high yield with high stability, making them the most desirable for selection. In contrast, landraces 9, 13, and 19 had above-average yield but low stability, reducing their suitability. Landraces 7 and 21 performed below the mean and were also highly unstable, marking them as the least favorable candidates.

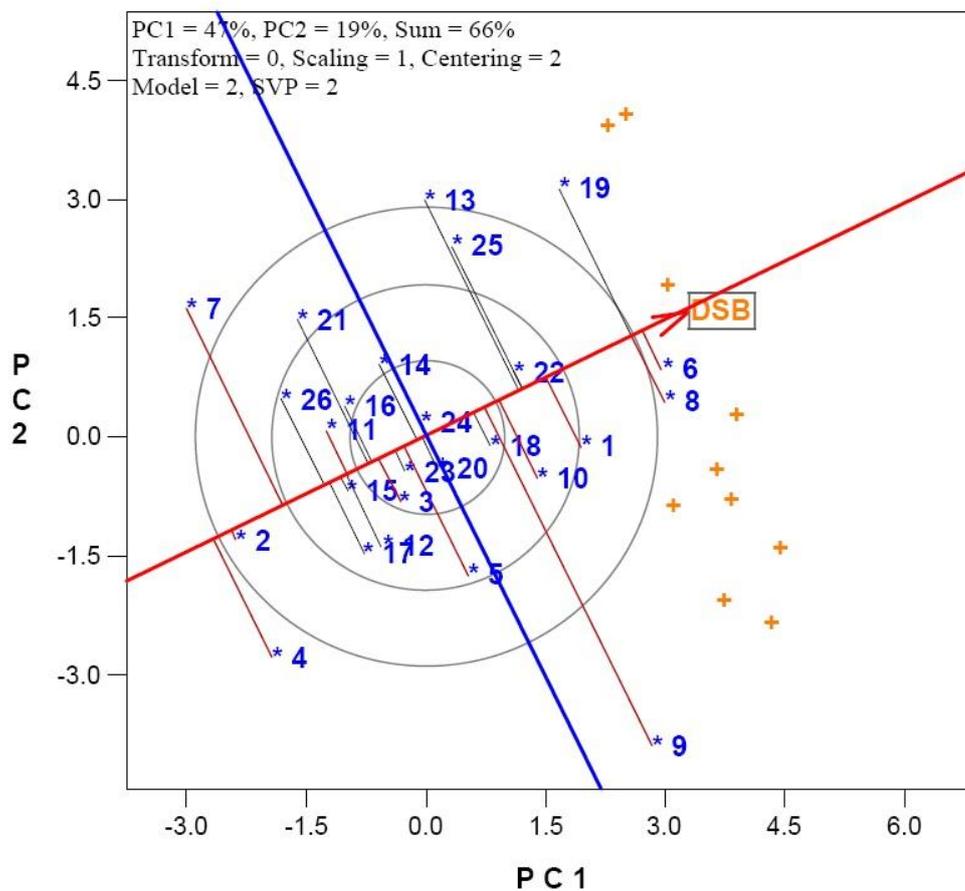


Figure 5. Examining the performance of/at fenugreek dry shoot biomass (DSB).

Discussion

The narrow genetic base of most cultivated fenugreek varieties underscores the urgent need to identify landraces with broader adaptive and yield-enhancing potential. The present study revealed substantial genetic variation, confirming earlier evidence of wide diversity within fenugreek germplasm (Meena *et al.* 2021; Roba Mohammed 2024). Importantly, the genotype-by-trait biplot captured both additive and crossover genotype \times trait interactions, highlighting the complexity of trait expression across landraces. Such interactions alter landrace rankings across traits and environments, complicating selection but also revealing opportunities to exploit specific strengths of individual landraces. This complexity is common in outcrossing and multipurpose crops, as reported for *Onobrychis* (Sabaghnia *et al.* 2024b) and safflower (Shekari *et al.* 2025), and must be explicitly considered in fenugreek breeding programs targeting resilience in semi-arid systems. A main outcome of this study was the identification of distinct trait clusters. For instance, branching and pod-related traits (number of lateral branches, middle leaf width, lateral leaf length, lateral leaf width, lateral leaf area, and pods per plant) grouped around the landrace 9 (Tehran-II), suggesting that vegetative architecture strongly supports

reproductive output. Physiologically, increased branching and leaf area enhance light interception and assimilate supply, which directly supports pod set and filling. In contrast, landraces 6 (Sarab) and 8 (Tabriz-II) were associated with the shoot biomass and middle leaf area, traits more reflective of vegetative growth capacity. Leaf area in particular serves as a proxy for photosynthetic potential, and its strong association with biomass aligns with the well-established role of canopy size in carbon assimilation (Hassan *et al.* 2025). Meanwhile, the landrace 19 (Ardabil), which excelled in chlorophyll traits, reflects a different adaptive strategy, maximizing photosynthetic pigment concentration rather than structural expansion. This divergence in trait associations demonstrates that fenugreek landraces may follow multiple physiological pathways to achieve higher biomass or reproductive success, a finding highly relevant for tailoring ideotypes to contrasting environments.

Contradictions in the literature about the relative importance of traits such as leaf area versus plant height and branching (Azizi *et al.* 2025; Hassan *et al.* 2025) can be understood in this light. Environments with ample radiation and limited water may favor compact plants with efficient pigment content, while environments with lower radiation but sufficient water may benefit from broader canopy structures that capture more light. This implies that the ideotype for fenugreek is not universal but should be context-specific: fodder-oriented ideotypes are landraces with large leaf area and high vegetative biomass (e.g., Sarab, Tabriz-II), grain yield ideotypes are landraces with strong branching and pod production capacity (e.g., Tehran-II), and stress-resilient ideotypes are landraces with high pigment efficiency or conservative growth forms (e.g., Ardabil). Another key insight lies in trait correlations. The strong positive association between shoot biomass and plant height suggests that height remains an important determinant for biomass. However, the weak relationship between chlorophyll traits and branching/pod traits suggests partial independence between physiological efficiency and architectural development. This independence is advantageous in breeding, as it allows for pyramiding of traits from different physiological domains; combining landraces with high pigment efficiency and those with strong branching could create hybrids with synergistic advantages. Conversely, the lack of negative correlations in this dataset is notable, as it suggests that trade-offs among measured traits are less constraining in fenugreek than in many crops. This could explain fenugreek's adaptability to diverse production systems, from fodder to spice cultivation.

The ideal genotype analysis reinforced the importance of combining performance with stability. Landraces 6 (Sarab) and 8 (Tabriz-II) not only excelled in biomass but also exhibited high stability, marking them as robust candidates for commercial release. This stability is particularly crucial for the role of fenugreek in marginal environments, where fluctuating conditions can penalize unstable landraces. By contrast, landraces such as 9, 13, and 19, despite showing high mean yield, displayed

high changeability, raising concerns about their adaptability. This illustrates the importance of integrating both mean performance and stability metrics in selection decisions, an issue echoed in recent ideotype studies of legumes (Climent *et al.* 2024). From a breeding perspective, the most discriminative and representative traits, such as middle leaf area, lateral leaf length, number of lateral branches, pods per plant, and plant height, seem to be practical selection criteria. Their dual role in differentiating landraces and capturing typical fenugreek phenotypes makes them valuable for early-generation selection. In contrast, chlorophyll traits, while biologically meaningful, may be better reserved as secondary criteria or for stress-focused breeding. A strategic approach could therefore involve using morphological traits as primary screening tools, followed by targeted physiological assessments to refine selections for stress adaptation or resource-use efficiency.

Finally, the integration of multivariate tools such as biplot analysis with physiological reasoning enables a more nuanced understanding of fenugreek improvement. Rather than searching for a single “superior” genotype, breeders can design ideotypes tailored to specific production objectives, fodder, grain yield, or dual-purpose, and target environments. Moreover, the absence of strong trade-offs among key traits in this study suggests that fenugreek ideotypes can potentially combine favorable morphological and physiological attributes without major compromise. The next step will be to validate these findings in multi-environment trials and integrate genomic approaches to accelerate the development of robust, high-performing fenugreek cultivars. This study demonstrated the utility of genotype-by-trait biplot analysis in fenugreek, which offers an integrative and visual tool to identify superior landraces and key traits beyond the scope of conventional analyses. Also, it delivered new insights into trait associations, revealing both synergistic and trade-off relationships, while distinguishing between discriminating and representative traits useful for selection. Finally, the study defined fenugreek ideotypes tailored to specific breeding objectives such as high grain yield, high biomass, or dual-purpose use, and linked these ideotypes to sets of morphological and agronomic traits. In practical terms, the findings indicate that breeders can focus on a smaller set of reliable traits to enhance selection efficiency, identify promising landraces for hybridization, and develop ideotype-oriented strategies for crop improvement. Finally, the study adds contextual novelty by extending advanced biplot methodology to an underutilized yet valuable legume, positioning fenugreek more prominently within the broader framework of functional food crop breeding.

Conclusion

This study revealed substantial genetic diversity among Iranian fenugreek landraces, with significant variation in key morphological and physiological traits influencing yield and biomass. The landrace

× trait biplot analysis identified positive associations between shoot biomass, plant height, leaf area, and branching traits, highlighting their importance for selection. Landraces 6 (Sarab) and 8 (Tabriz-II) demonstrated superior performance and stability, making them promising candidates for breeding and commercial release. The findings emphasize the value of integrating genotype × trait interaction analysis in fenugreek improvement programs.

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Conflict of Interest

The authors declare that they have no conflict of interest with any individual or organization concerning the subject of this article.

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