RESEARCH ARTICLE Legume Research- An International Journal, Volume 49 Issue 6: 934-942 (June 2026)

Identification of a Novel Genotype with Determinate Growth Habit in Fenugreek (*Trigonella foenum-graecum* L.)

Ambika Baldev Gaikwad[1], Sheel Yadav[1], Neelam Shekhawat[2], Sunil Gomashe[3], Ratna Kumari[1], Vinod Kumar Sharma[1], Dhirendra Singh[4], D.K. Gothwal[4], Narendra Kumar Gupta[4]

10.18805/LR-5616

ABSTRACT

Background: Fenugreek (*Trigonella foenum-graecum* L.), also known as methi, is a crop with immense economic and therapeutic importance. It is widely used for human consumption, both as a leafy vegetable and as a seed spice. Crop improvement in fenugreek entails characterization of the germplasm for traits of agronomic importance. Novel sources of variation are required to be identified for this purpose.

Methods: A novel genotype with determinate growth habit has been evaluated with different genotypes of fenugreek at four different locations for various quantitative and qualitative traits. Descriptive statistical analysis was performed for these traits to identify trait variation existing in the germplasm.

Result: Highest variation was observed for the trait of number of primary branches per plant while least variation was observed for days to 50% flowering. The qualitative traits demonstrated not much variation as these are oligogenic in nature. Amongst the accessions, a genotype, UM-370 was identified to possess the determinate growth habit. This genotype was found to be earlier flowering than the previously known determinate genotype, RMt-305. UM-370 was characterized using DNA-based molecular markers to establish its distinctness. This is the first report on identification of UM-370 as a determinate genotype in fenugreek. The identified genotype can be utilized to breed for the determinacy trait in fenugreek.

Key words: Determinate, Fenugreek, Molecular markers, Plant growth habit, Qualitative, Quantitative traits.

INTRODUCTION

Fenugreek (*Trigonella foenum-graecum* L.) or methi, is a crop with significant economic and medicinal value (Mehrafarin *et al*., 2011; Choudhary *et al*., 2024; Yadav *et al*., 2024). It belongs to the Fabaceae family. It is a multi-purpose crop, where leaves are consumed as vegetable and as fodder for livestock (Singh *et al*., 2025; Parveen and Samyuktha, 2026). For medicinal purposes, the leaves, seeds and sometimes the whole plant is used (Sinskaja, 1961). These applications have contributed to its popularity, leading to its cultivation in nearly every region of the world for centuries (Sinskaja, 1961). However, India is the largest producer of fenugreek in the world with the states of Madhya Pradesh, Haryana, Rajasthan, Gujarat, Maharashtra, Uttar Pradesh, Punjab, Bihar and Andhra Pradesh, as the major fenugreek producers (Meena *et al*., 2018). In India, it is one of the oldest used spices, seeds (vernacularly called “methi dana”) of which are rampantly used as a condiment or flavouring agent in many Indian cuisines (Srinivasan, 2014). Despite its economic importance, fenugreek remains largely underutilized and neglected as compared to other crops, and its potential in medicine remains underexplored. The germplasm of a species serves as a valuable resource by providing plant breeders unique opportunities to develop new crop varieties. The successful application of germplasm collections for enhancing crop productivity depends on comprehending the extant genetic diversity (Mondal *et al*., 2023). India being the primary or main fenugreek producer, has a rich diversity in fenugreek germplasm for many morphological traits like plant height, growth pattern, growth habit, yield related traits, *etc*. Many of these traits have been investigated to gauge the genetic diversity within fenugreek germplasm (Maloo *et al*., 2020; Roba and Mohammed, 2024). A cultivar demonstrates significant variation in growth performance when grown in different environments, a phenomenon referred to as genotype-environment interaction (GEI) (Dos *et al*., 2003). The phenotype is the

[1]ICAR-National Bureau of Plant Genetic Resources, New Delhi-110 012, India.
[2]ICAR-National Bureau of Plant Genetic Resources, Regional Station, Jodhpur-342 003, Rajasthan, India.
[3]ICAR-National Bureau of Plant Genetic Resources, Regional Station, Akola-444 104, Maharashtra, India.
[4]Sri Karan Narendra Agricultural University, Jobner-303 329, Rajasthan, India.

Corresponding Author: Ambika Baldev Gaikwad, ICAR-National Bureau of Plant Genetic Resources, New Delhi-110 012, India. Email: ambikabg@gmail.com

How to cite this article: Gaikwad, A.B., Yadav, S., Shekhawat, N., Gomashe, S., Kumari, R., Sharma, V.K., Singh, D., Gothwal, D.K. and Gupta, N.K. (2026). Identification of a Novel Genotype with Determinate Growth Habit in Fenugreek (*Trigonella foenum-graecum* L.). *Legume Research.* **49(6):** 934-942. doi: 10.18805/LR-5616.

Submitted: 02-12-2025 **Accepted:** 05-03-2026 **Online:** 30-03-2026

result of the interplay between genetic factors, environmental conditions and their interaction. In order to estimate GEI, it is therefore important to grow cultivars across various agro-climatic zones which allows selection of superior genotypes which can be subsequently utilized for varietal development.

The cultivated legumes acquired the determinate growth habit where the vegetative growth terminates into a cluster of flowers, during domestication as a component of the "domestication syndrome". This type of growth habit is associated with several advantages like early and synchronous maturity, a compact plant type, and potentially higher per-plant yield (Avtar and Jhorar, 2002). Plants with determinate growth habit are compact and more amenable to mechanical harvesting and therefore are more desirable. Owing to these advantages, breeding for determinate growth habit remains a priority in crop improvement. The genetic control of the determinate growth habit has been deciphered in many legumes with the trait being under monogenic (pigeonpea) or digenic (chickpea) inheritance in most species (Kapoor and Gupta, 1991). The performance of RILs (recombinant inbred lines) derived from DT (determinate) × IDT (indeterminate) crosses of Indian bean depicted segregation for various traits like flowering time, plant height, number of racemes produced, and several important yield-related traits, likely due to co-localization of the genes governing the growth habit and these traits (Megha *et al*., 2023). Identifying the genetic loci governing the determinate growth habit would therefore help in developing desirable plant types by allowing improvement of associated traits. Typically like most legumes, fenugreek has an indeterminate growth habit where the plants grow continuously at the shoot apex and they keep producing new shoots, flowers and seed pods (Avtar and Jhorar, 2002). However, a few determinate type of fenugreek varieties have also been identified or developed (Chaudhary and Singh, 2001; Avtar and Jhorar, 2002; Choudhary, 2003). Notable amongst these is RMt-305 which is a determinate variety that was developed from RMt-1 using ethyl methane sulphonate (EMS) mutagenesis (Chaudhary and Singh, 2001). Identification of new determinate genotypes would allow broadening of the genetic base for genetic improvement for the trait.

Keeping in view, the importance of morphological characterization for trait discovery, the present study was undertaken to morphologically characterize a set of 30 different fenugreek germplasm lines across four different locations. Through this analysis, we identified a novel determinate genotype. The genotype identified has not been previously reported as a determinate genotype. The novelty of this genotype as a genotype with determinate growth habit was established using DNA-based molecular markers. Based on the profiles obtained, it could be concluded that the identified genotype is different from the previously reported determinate genotype. The identification of this genotype would facilitate breeding for determinate plant type in fenugreek.

MATERIALS AND METHODS

Plant material and experimental design

A total of 300 accessions of fenugreek were grown in randomized block design (RBD) during *Rabi* 2023 for germplasm characterization at ICAR-National Bureau of Plant Genetic Resources, Pusa Farm, New Delhi (28°34′ North and 76°50′ East) (unpublished data). A genotype UM-370 was identified as a novel genotype with determinate growth habit. This genotype was shorter in height and the shoot apex terminated into inflorescence, typical of the determinate growth habit. In the following year (*Rabi* 2024) a set of thirty accessions, including UM-370 were evaluated in augmented block design (ABD) under natural field conditions, at four different locations, namely ICAR-NBPGR, Pusa Farm, New Delhi (28°34′ North and 76°50′ East); ICAR-NBPGR, Issapur Farm, New Delhi (28°63′ North and 77°16′ East); ICAR-NBPGR Regional Station, Jodhpur (26°18′ North and 73°00′ East) and ICAR-NBPGR Regional Station, Akola (20°43′ North and 77°04′ East). These accessions were selected based on the diversity for various traits recorded and few of these were cultivated varieties (Table 1). Each accession was grown in two rows of 2 m length each. The row-to-row distance was maintained at 45 cm and the plant to plant spacing was maintained at 15 cm. Standard agronomic practices were followed throughout the crop growth period (Bhutia *et al.,* 2018). The check varieties included the varieties Hisar Sonali and RMt-1.

Phenotypic evaluation

The germplasm was characterized for various qualitative and quantitative traits following the DUS (Distinctiveness, Uniformity and Stability) guidelines (https://plantauthority.gov.in). The data was recorded on five randomly selected plants of each accession (excluding the border plants). The quantitative traits included days to 50% flowering (DTF);

Table 1: The list of the thirty genotypes of fenugreek characterized in the present study.

S. no.	Genotype	S. no.	Genotype
1	UM-37	16	UM-303
2	UM-64	17	UM-326
3	Co-1	18	UM-340
4	Rajendra kanti	19	UM-358
5	UM-82	20	UM-370
6	AM-2	21	UM-297
7	UM-90	22	UM-368
8	Hisar Sonali (HS)	23	UM-298
9	UM-211	24	UM-254
10	HM-444	25	UM-209
11	Pusa early bunching	26	UM-327
12	RMt-1	27	UM-92
13	UM-231	28	UM-341
14	RMt-305	29	JP-4
15	UM-282	30	UM-378

plant height (PH); number of primary branches per plant (PB); number of pods per plant (PP); pod length (PL) and 1000 seed weight (TSW). The qualitative characters included shape of leaf blade, plant growth pattern, pod curvature and plant growth habit.

Statistical analysis

The phenotypic data collected was used for further analysis. XLSTAT (Addinsoft, Paris) software (2020) was utilized to construct boxplots, correlation coefficient and principal component analysis (PCA). To perform hierarchical clustering using the UPGMA method, we used the 'dendextend' and 'factoextra' packages using the R programming language. ANOVA (Analysis of Variance) analysis was carried out for the trait data across the four different locations using https://www.statskingdom.com.

Molecular marker based validation of UM-370

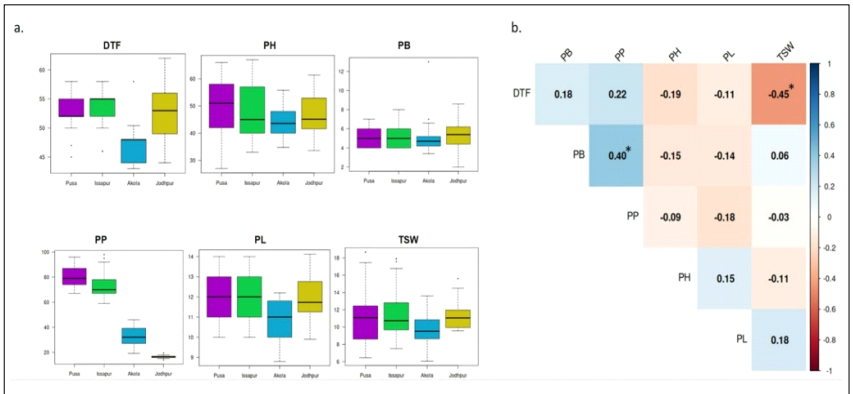
The genotype, UM-370 was identified as a new determinate type of fenugreek. As molecular markers are the ultimate line of evidence to establish the distinctness of different genotypes, we used SSR markers to establish the distinctness of UM-370 from the earlier identified determinate variety, RMt-305. PCR amplification was performed for these two genotypes along with the indeterminate varieties Hisar Sonali and RMt-1 as check samples. The PCR reactions were performed in a total volume of 20 µl, with the reaction mixture consisting of 1X PCR buffer, 2.5 mM $MgCl_2$, 1 µM primer, 0.2 mM of each dNTPs, 1 U Taq DNA polymerase (NEB) and 15 ng template DNA. The standard PCR amplification conditions were used (Gaikwad *et al.*, 2025). The amplified products were resolved and visualized through EtBr staining of 3% metaphor agarose gels.

RESULTS AND DISCUSSION

A large variability was observed for the traits studied, with highest variability for the trait PB (CV of 25%). The PB values ranged from 3.9 to 8.4 for the pooled data. This is in agreement with previous report where the PB was reported to range from 2.3 to 7.5 across 245 different fenugreek genotypes (Sharma and Sastry, 2008). The trait DTF was least variable (CV of 6.5%). The DTF ranged from a minimum of 45.6 to 58. Highest variation between the four locations was observed for the trait PP with the genotypes at Jodhpur with a substantially lesser number of average pods/plant compared to the average PP for genotypes grown at other locations (Fig 1a, Table 2). ANOVA analysis depicted a significant ($p<0.05$) difference in means for the traits of DTF, PP, PL and TSW depicting higher trait variability across the four different locations (Table 3).

Correlation analysis revealed a significant ($p\leq0.05$) negative correlation (-0.45) between TSW and DTF; and a significant ($p\leq0.05$) positive correlation between PB and PP (0.40) (Fig 1b). The PCA analysis indicated that the first two principal components explained a cumulative variance of more than 50% (Table 4, Fig 2a). The PC1 possessed the highest eigenvalue and % variance suggesting that it accounts for the largest variation in the dataset examined. The traits DTF and PP possessed the highest positive eigenvalues for the PC1 indicating their significant influence in governing the overall variability among the fenugreek genotypes. In the correlation circle for PCA, the varied contribution of each of the traits towards the PC1 and 2 is depicted in the form of variation in vector lengths (Fig 2b). The traits PH and PL possessed the least vector lengths. In the scatter plot, the proximity of the vectors depicts presence of a strong correlation between the variables. Such vectors include PP and PB. Overall, the results of this analysis, concurred well with the correlation analysis.

Cluster analysis revealed the presence of two distinct clusters with cluster I containing twelve genotypes and cluster II possessing eighteen genotypes (Fig 3). Cluster I was further divided into two sub-clusters and cluster II was also similarly divided into two sub-clusters with only one genotype, CO 1 in one of the subclusters.



a) Box plot representing variation for different quantitative traits like DTF: Days to 50% flowering; PH: Plant height; PB: Number of primary branches per plant; PP: Number of pods per plant; PL: Pod length and TSW: 1000 seed weight. b) Correlation coefficient matrix plotted for the traits.

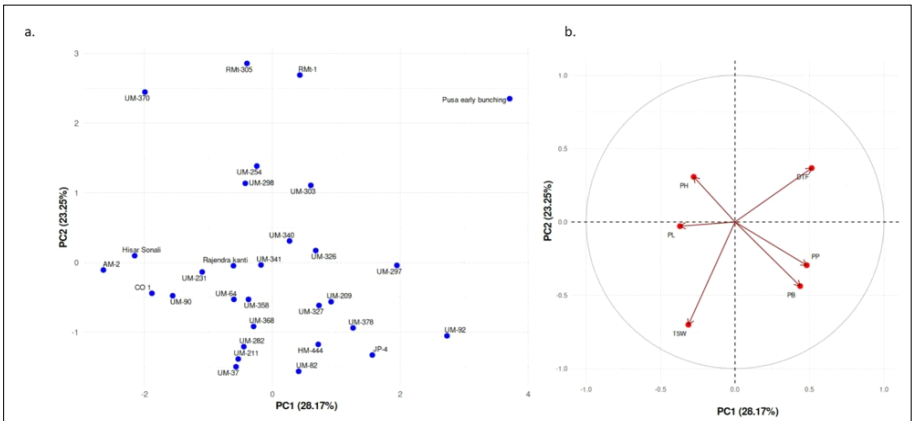
Fig 1: Trait variability in fenugreek germplasm.

The genotypes were analysed for the qualitative characters (Table 5). This includes the plant growth habit, shape of leaf blade (basal and apex) at first primary branch axis and first pod axis, shape of leaf blade for fully grown terminal leaf, plant growth pattern and curvature of the pods. There was little variation for these traits across the four locations, as qualitative traits show little environmental variation. This is because they are oligogenic in nature

Table 2: Statistical analysis of various quantitative traits in fenugreek, with data recorded at four different locations (n=5).

Parameters	DTF				PH			
	Pusa	Issapur	Akola	Jodhpur	Pusa	Issapur	Akola	Jodhpur
Minimum	45	46	43	44	27	33	34.8	33.6
Frequency of minimum	1	2	2	2	1	2	1	1
Maximum	58	58	58	62	66	67	55.8	61.3
Frequency of maximum	1	5	1	1	1	1	1	1
Mean	52.4	53.7	47.1	53.0	49.7	48.6	44.5	46.6
Standard deviation (SD)	2.7	3.2	3.05	4.5	11.05	10.2	5.7	8.2
Coefficient of variation (CV) (%)	5.1	6.1	6.4	8.4	22.2	21.1	12.9	17.7
Parameters	PB				PP			
	Pusa	Issapur	Akola	Jodhpur	Pusa	Issapur	Akola	Jodhpur
Minimum	4	4	3.4	2	67	59	19.2	14
Frequency of minimum	8	12	2	1	1	1	1	1
Maximum	7	8	13	8.6	96	98	46	19.4
Frequency of maximum	3	1	1	1	1	1	1	1
Mean	5.2	5.0	5.01	5.3	80.3	73	32.8	16.4
Standard deviation (SD)	0.96	1.09	1.7	1.4	8.2	9.8	6.9	1.08
Coefficient of variation (CV) (%)	18.4	21.8	34.1	27.3	10.3	13.4	21.2	6.5
Parameters	PL				TSW			
	Pusa	Issapur	Akola	Jodhpur	Pusa	Issapur	Akola	Jodhpur
Minimum	10	10	8.8	9.9	6.4	7.5	6.0	9.5
Frequency of minimum	3	4	2	1	1	1	1	1
Maximum	14	14	12.2	14.1	18.6	17.8	13.6	15.6
Frequency of maximum	2	2	1	1	1	1	1	1
Mean	12	12	10.9	11.9	11.2	11.6	9.7	11.3
Standard deviation (SD)	1.1	1.1	0.9	1.0	3.1	2.8	1.7	1.6
Coefficient of variation (CV) (%)	9.5	9.5	8.4	8.6	27.7	24.4	18.1	14.8

DTF: Days to 50% flowering; PH: Plant height; PB: Number of primary branches per plant; PP: Number of pods per plant; PL: Pod length and TSW: 1000 Seed weight.



a) PCA biplot of genotypes in relation to PC1 and PC2. b) Correlation circle of PCA with various traits shown as vectors.

Fig 2: Principal component analysis (PCA) for the traits.

being controlled by a single gene or a small number of genes. Notably, for a few categories no observation was recorded. For instance, none of the genotypes were found to possess rounded shape of the apex of leaf blade on first pod axis. For pod curvature, only Hisar Sonali was observed to possess strongly curved pods with the remaining genotypes having pods with moderate curvature. For the trait of plant growth habit, only two genotypes were observed with the determinate growth habit, namely RMt-305 and UM-370.

Table 3: ANOVA analysis for the different traits across four different locations.

Source	df	Sum of square (SS)	Mean square (MS)	F statistic	P-value
		DTF			
Between groups	3	808.62	269.54	23.18	0.000
Within groups	115	1336.98	11.63		
Total	118	2145.60			
		PH			
Source	df	Sum of square (SS)	Mean square (MS)	F statistic	P-value
Between groups	3	472.69	157.56	1.91	0.13
Within groups	116	9521.43	82.08		
Total	119	9994.13			
		PB			
Source	df	Sum of square (SS)	Mean square (MS)	F statistic	P-value
Between groups	3	2.08	0.69	0.38	0.76
Within groups	116	208.36	1.80		
Total	119	210.44			
		PP			
Source	df	Sum of square (SS)	Mean square (MS)	F statistic	P-value
Between groups	3	85977.83	28659.27	532.18	0.00
Within groups	116	6246.78	53.85		
Total	119	92224.61			
		PL			
Source	df	Sum of square (SS)	Mean square (MS)	F statistic	P-value
Between groups	3	25.79	8.59	7.58	0.00011
Within groups	116	131.43	1.13		
Total	119	157.22			
		TSW			
Source	df	Sum of square (SS)	Mean square (MS)	F statistic	P-value
Between groups	3	61.50	20.50	3.46	0.018
Within groups	116	687.03	5.92		
Total	119	748.54			

Where, DTF: Days to 50% flowering; PH: Plant height; PB: Number of primary branches per plant; PP: Number of pods per plant; PL: Pod length and TSW: 1000 seed weight.

Table 4: Principal component analysis (PCA) based eigen vectors.

Parameter	PC1	PC2	PC3	PC4	PC5	PC6
Eigenvalue	1.81	1.31	0.97	0.89	0.58	0.41
% of variance	30.22	21.95	16.20	14.89	9.80	6.92
Cumulative (%)	30.22	52.17	68.37	83.27	93.07	100
DTF	0.51	0.36	-0.07	0.43	0.01	0.63
PH	-0.27	0.30	0.79	-0.27	0.10	0.32
PB	0.43	-0.43	0.31	0.08	0.70	-0.13
PP	0.48	-0.29	0.41	-0.02	-0.69	-0.13
PL	-0.36	-0.02	0.28	0.85	-0.06	-0.22
TSW	-0.31	-0.70	-0.07	0.02	-0.08	0.62

The variance contribution (%) from the principal component (PC) axes is depicted. DTF: Days to 50% flowering; PH: Plant height; PB: Number of primary branches per plant; PP: Number of pods per plant; PL: Pod length and TSW: 1000 seed weight.

Table 5: Characterization and classification of fenugreek genotypes for qualitative traits.

Trait	Categories	Genotypes
Basal shape of first leaf blade	Acute	UM-37, UM-64, CO 1,AM-2, UM-211, HM-444, Pusa early bunching, UM-231, UM-282, UM-326, UM-254, UM-92, UM-341, JP-4, UM-378
	Obtuse	RMt-1, RMt-305, Hisar Sonali, Rajendra kanti, UM-82, UM-90, UM-303, UM-340, UM-358, UM-370, UM-368, UM-298,UM-209, UM-327
	Rounded	UM-297
Apex shape of first leaf blade	Obtuse	RMt-1, RMt-305, UM-37, UM-64,CO 1, UM-82, UM-90, UM-211, Pusa early bunching, UM-231, HM-444, UM-303, UM-326, UM-340, UM-358, UM-368, UM-298, UM-254,UM-92, UM-341, JP-4,UM-378
	Rounded	Hisar Sonali, Rajendra kanti, AM-2, UM-282, UM-370,UM-297, UM-209, UM-327
Basal shape of leaf blade on first primary branch axis	Acute	Hisar Sonali, RMt-305,UM-37, CO 1, UM-82, UM-211, HM-444, Pusa early bunching, UM-282, UM-303, UM-326, UM-340, UM-297, UM-368, UM-298, UM-254, UM-92, UM-341, JP-4, UM-378
	Obtuse	Rajendra kanti, RMt-1, UM-64, AM-2, UM-90, UM-231, UM-358, UM-370, UM-209, UM-327
Apex shape of leaf blade on first primary branch axis	Acute	UM-37,CO 1,UM-82, UM-211,HM-444, Pusa early bunching, UM-282, UM-303, UM-326, UM-340, UM-297, UM-368, UM-298, UM-254, UM-92, UM-341, JP-4, UM-378
	Obtuse	UM-64, Rajendra kanti, AM-2,UM-90, UM-231, UM-358, UM-370, UM-209, UM-327
	Rounded	Hisar Sonali, RMt-1, RMt-305
Basal shape of leaf blade on first pod axis	Acute	UM-37, UM-64, UM-90, CO 1, UM-82, HM-444, UM-231, UM-303, UM- 326, UM-340, UM-358, UM-368, UM-298, UM-254, UM-92, UM-341, JP-4, UM-378, UM-211
	Obtuse	Rajendra kanti, AM-2, Hisar Sonali, RMt-1, RMt-305, Pusa early bunching, UM-282,UM-370, UM-297,UM-209, UM-327
Apex shape of leaf blade on first pod axis	Acute	RMt-1, RMt-305, Rajendra kanti, Hisar Sonali,UM-37, CO 1, UM-82, UM-211, HM-444, Pusa early bunching, UM-282, UM-303, UM-326, UM-340, UM-297, UM-368, UM-298, UM-254, UM-92, UM-341, JP-4,UM-378
	Obtuse	UM-64, AM-2, UM-90, UM-231, UM-358, UM-370, UM-209, UM-327
	Rounded	
Basal shape of leaf blade on fully grown terminal leaf	Acute	UM-37,CO 1, Rajendra kanti, UM-82, Hisar Sonali, UM-211, HM-444, Pusa early bunching, RMt-1, RMt-305, UM-282, UM-303, UM-326, UM-340, UM-297, UM-368, UM-298, UM-254, UM-92, UM-341, JP-4, UM-378
	Obtuse	UM-64,AM-2,UM-90,UM-231,UM-358,UM-370, UM-209, UM-327
Apex shape of leaf blade on fully grown terminal leaf	Acute	UM-37, CO 1, Rajendra kanti, UM-82, Hisar Sonali, UM-211, HM-444, Pusa early bunchy, RMt-1, RMt-305, UM-282, UM-303, UM-326, UM-340, UM-92,UM-341,UM-297,UM-368,UM-298,UM-254,UM-378, JP-4
	Obtuse	UM-64, UM-209, UM-327, AM-2, UM-90, UM-231, UM-358, UM-370
Plant growth pattern	V type	UM-37, UM-64, CO 1, Rajendra kanti, UM-82, AM-2, UM-90, HM-444, RMt-1, RMt-305, UM-303, UM-326, UM-340, UM-358, UM-370, UM-297, UM-368, UM-298, UM-254, UM-209, UM-327, UM-92, JP-4, UM-378
	U type	Hisar Sonali, UM-211, Pusa early bunching, UM-231, UM-282, UM-341
Pod curvature	Moderately curved	UM-37, UM-64, CO 1, Rajendra kanti, UM-82, AM-2, UM-90, HM-444, RMt-1, RMt-305, UM-303, UM-326, UM-340, UM-358, UM-370, UM-297, UM-368, UM-298, UM-254, UM-209, UM-327, UM-92, JP-4, UM-378, UM-211, Pusa early bunching, UM-231, UM-282, UM-341
	Strongly curved	Hisar Sonali
Plant growth habit	Determinate	RMt-305, UM-370
	Indeterminate	UM-37, UM-64, CO 1, Rajendra kanti, UM-82, AM-2, UM-90, HM-444, RMt-1, UM-303, UM-326, UM-340, UM-358, UM-297, UM-368, UM-298, UM-254, UM-209, UM-327, UM-92, JP-4, UM-378, UM-211, Pusa early bunching, UM-231, UM-282, UM-341, Hisar Sonali

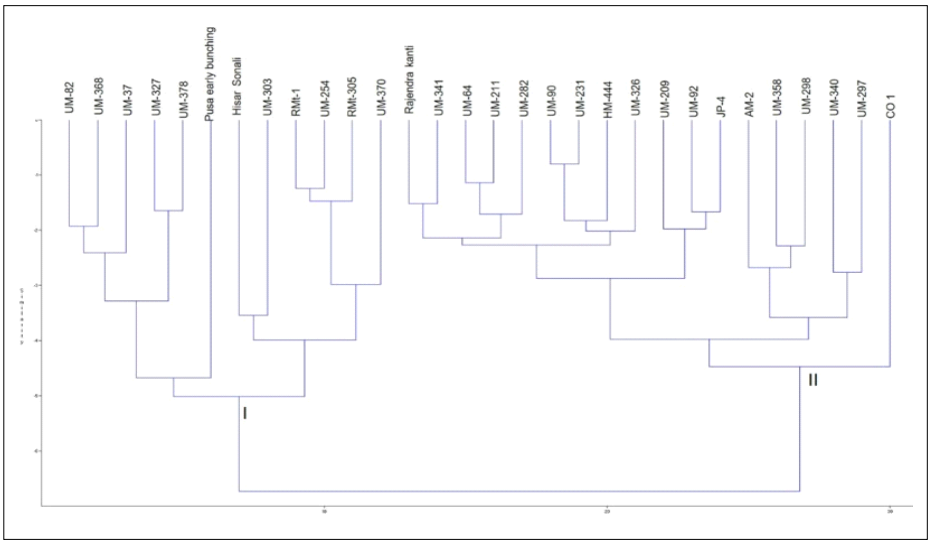


Fig 3: Dendrogram representing clustering of thirty genotypes based on the different quantitative traits.

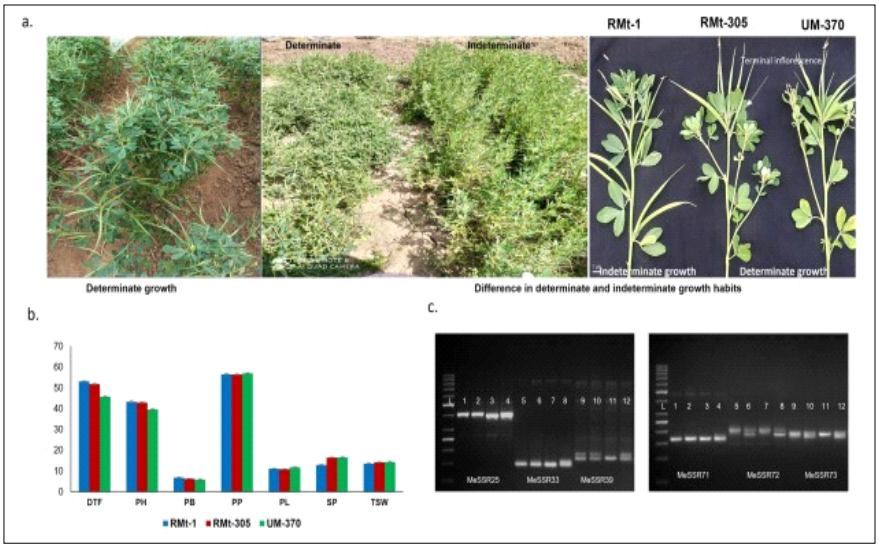


Fig 4: a) Representative field images of plants depicting the difference in determinate and indeterminate growth habit. b) Comparative performance of the three genotypes for various traits like DTF: Days to 50% flowering; PH: Plant height; PB: Number of primary branches per plant; PP: Number of pods per plant; PL: Pod length; SP: Seed yield per plant and TSW: 1000 seed weight. c) DNA based molecular profiling of four different accessions (in sets of 4) using various SSR markers. L: 100 bp plus DNA ladder; 1: Hisar Sonali; 2: RMt-1; 3: RMt-305; 4: UM-370. MeSSRs (Methi Simple Sequence Repeat) markers.

Amongst the genotypes, a determinate genotype, UM-370, was confirmed through phenotypic observations, at all the four locations. This genotype was earlier maturing and shorter than the other determinate variety, RMt-305 (Fig 4a and b). It is a local collection from village Manpura, in Nagaur, Rajasthan. The molecular profiling of this genotype with SSR (Simple Sequence Repeat) markers confirmed its distinctness from RMt-305 (Fig 4c). SSR markers have the advantage of identifying both genic and intergenic variation by virtue of their presence in either regions of the genome and they are widely used for genetic diversity analysis and gene mapping or tagging (Jethra *et al.,* 2020; Maloo *et al.,* 2023). The SSR markers which are identified to be polymorphic between the determinate and indeterminate genotypes in the present study could potentially indicate allelic variation for the genomic region governing the plant growth habit in fenugreek. Based on the obtained SSR profiles, UM-370 was identified to be a genotype different from the previously known determinate variety RMt-305. The identification of a novel determinate genotype would provide unique opportunities for breeding for determinate growth habit in fenugreek. In the absence of trait variability for plant growth habit in fenugreek, identification of a novel determinate genotype would broaden the genetic base for development of determinate varieties in fenugreek. Since the determinate genotypes

are earlier maturing, they are capable of escaping the peak heat or drought periods, thereby ensuring stable yields under the rapidly changing environmental conditions.

CONCLUSION

In this study, we characterized the fenugreek germplasm for various quantitative and qualitative traits, at four different locations. Substantial variation existed for the quantitative traits while not much variation was observed for the qualitative traits. Highest variation was observed for the number of primary branches per plant while least variation was observed for the days to 50% flowering. A determinate genotype, UM-370 was identified and its distinctness from the previously known determinate genotype, RMt-305 was established using SSR markers. This genotype can be used for breeding fenugreek for the determinacy trait. The genotype identified would broaden the genetic base for fenugreek improvement in the current scenario of rapidly changing climate as the determinate genotypes are higher yielding and with synchronous maturity.

ACKNOWLEDGEMENT

The authors acknowledge the facilities provided at ICAR-National Bureau of Plant Genetic Resources.

Funding

The work was supported by the funding provided by ICAR-Consortium Research Platform on Genomics (Project number 1007341).

Author contributions

Ambika Baldev Gaikwad conceptualized the study, procured the grants, drafted and edited the manuscript. Sheel Yadav, Neelam Shekhawat, Sunil Gomashe, Ratna Kumari, Vinod Kumar Sharma carried out the experiments, Sheel Yadav drafted the manuscript, Dhirendra Singh and D.K. Gothwal provided the material, Narendra Kumar Gupta edited the manuscript. All authors have read and approved the final manuscript.

Conflict of interest

All authors declared that there is no conflict of interest.

REFERENCES

Avtar, R. and Jhorar, B.S. (2002). Inheritance of determinate growth habit in fenugreek (*Trigonella foenum-graecum* L.). *Journal of Spices and Aromatic Crops*. **11(2):** 146-147.

Bhutia, P.H. and Sharangi, A.B. (2018). Influence of dates of sowing and irrigation scheduling on phenology, growth and yield dynamics of fenugreek (*Trigonella foenum-graecum* L.). *Legume Research-An International Journal*. **41(2):** 275-280. doi: 10.18805/LR-3432.

Chaudhary, A.K. and Singh, V.V. (2001). An induced determinate mutant in Fenugreek (*Trigonella foenum-graecum* L.). *Journal of Spices and Aromatic Crops*. **10:** 51-53.

Choudhary, A.K. (2003). Outcrossing behaviour in fenugreek (*Trigonella foenum-graecum* L.). *Indian Journal of Genetics and Plant Breeding*. **63(02):** 178-178.

Choudhary, S., Singh, R., Ravi, Y., Gena, C.B., Singh, D., Harisha, C.B., Meena, R.S., Meena, N.K., Chauhan, V.B.S., Kavan, Kumar, V., Versha, Kumari. and Verma A.K. (2024). Phytotherapeutic, Nutraceutical, Medicinal, and forage properties of fenugreek (*Trigonella foenum-graecum* L.): A comprehensive review. *Journal of Advances in Biology and Biotechnology*. **27:** 719-733.

Dos Santos, M.M., Gombert, A.K., Christensen, B., Olsson, L. and Nielsen, J. (2003). Identification of *in vivo* enzyme activities in the cometabolism of glucose and acetate by *Saccharomyces cerevisiae* by using 13C-labeled substrates. *Eukaryot Cell*. **2(3):** 599-608. doi: 10.1128/EC.2.3.599-608.2003.

Gaikwad, A.B., Yadav, S., Kumari, R., Maurya, W., Rangan, Singh, R. and Singh, G.P. (2025). Chromosome-scale genome assembly of *Trigonella corniculata* (L.) L. (Nagauri pan /Kasuri methi), an important spice. *Scientific Data*. **12:** 509 https://doi.org/10.1038/s41597-025-04858-4.

Jethra, G., Choudhary, S. and Sharma, V. (2020). Identification and characterization of microsatellite markers in fenugreek: An inter-family amplification. *Legume Research-An International Journal*. **43(5):** 611-616. doi: 10.18805/LR-4024.

Kapoor, R.K. and Gupta, S.C. (1991). Inheritance of growth habit in pigeonpea. *Crop Science*. **31(6):** 1456-1459.

Maloo, S.R., Sharma, R. and Soan, H. (2023). SSR based genetic diversity analysis in fenugreek (*Trigonella foenum-graecum* L.) genotypes. *Legume Research-An International Journal*. **46(3):** 307-311. doi: 10.18805/LR-4787.

Maloo, S.R., Sharma, R., Jain, D., Chaudhary, S. and Soan, H. (2020). Assessment of genetic diversity in fenugreek (*Trigonella foenum-graecum*) genotypes using morphological and molecular markers. *The Indian Journal of Agricultural Sciences*. **90(1):** 25-30.

Meena, R.S., Lal, G. and Meena, N.K. (2018). Effect of promising Indian varieties of fenugreek (*Trigonella foenum-graecum*) on yield attributed, yield and its economics. *Bhartiya Krishi Anusandhan Patrika*. **33(3):** 206-208. doi: 10.18805/BKAP118.

Megha, L.M., Mendapara, I., Kaushal, M., Patel, R. and Parmar, V. (2023). Performance of determinate and indeterminate RILs of Indian bean (*Lablab purpureus* L. Sweet). *The Pharma Innovation Journal*. **12(3):** 3573-3576.

Mehrafarin, A., Rezazadeh, S.H., Naghdi Badi, H., Noormohammadi, G.H., Zand, E. and Qaderi, A. (2011). A review on biology, cultivation and biotechnology of fenugreek (*Trigonella foenum-graecum* L.) as a valuable medicinal plant and multipurpose. *Journal of Medicinal Plants*. **10(37):** 6-24.

Mondal, R., Kumar, A. and Gnanesh, B.N. (2023). Crop germplasm: Current challenges, physiological-molecular perspective and advance strategies towards development of climate-resilient crops. *Heliyon*. **9(3).** doi: 10.1016/j.heliyon.2023.e12973.

Parveen Sumaiya, S. and Samyuktha, C. (2026). Spatial dynamics of fenugreek pests and their natural predators: Influence of environmental factors on aphid infestation and sustainable management. *Legume Research*. **49(5):** 876-882. doi: 10.18805/LR-5623.

Roba, R. and Mohammed, W. (2024). Genetic variability of fenugreek (*Trigonella foenum-graecum* L.) accessions from agroecological and morphoagronomic traits, Ethiopia. *Beverage Plant Research*. **4(1):** 1-11. doi: 10.48130/bpr-0024-0003.

Singh, R., Meena, R.S., Choudhary, S., Meena, N.K., Meena, R.D., Verma, A.K., Mahatma, M.K., Yathendranaik, R., Lal, S., Shekhawat, P.K. and Bhardwaj, V. (2025). Deciphering agronomic traits, biochemical components, and color in unique green-seeded fenugreek (*Trigonella foenum-graecum* L.) genotypes. *Frontiers in Nutrition*. **12:** 1542211. doi: 10.3389/fnut.2025.1542211.

Sinskaja, E. (1961). Flora of the Cultivated Plants of the U.S.S.R. XIII. In: *Perennial leguminous* Plants. Part I: Medicago, Sweet Clover, Fenugreek. Jerusalem: Israel Program for Scientific Translations.

Srinivasan, K. (2014). Antioxidant potential of spices and their active constituents. *Critical Reviews in Food Science and Nutrition.* **54(3):** 352-372. doi: 10.1080/10408398.2011.585525.

Yadav, S., Kumari, R., Rangan, P. and Gaikwad, A.B. (2024). Variability in genome size of *Trigonella foenum-graecum*, *Trigonella corniculata* and *Trigonella caerulea* as estimated by flow cytometry indicates complex evolutionary history of fenugreek. *Molecular Biology Reports*. **51:** 489. https://doi.org/10.1007/s11033-024-09417-5.