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The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2023; 12(7): 561-565

www.thepharmajournal.com
Received: 23-04-2023
Accepted: 29-05-2023

Himanshi
M.Sc. Scholar, College of Agriculture, Ummedganj, Agriculture University, Kota, Rajasthan, India

Khajan Singh
Assistant Professor, College of Agriculture, Ummedganj, Agriculture University, Kota, Rajasthan, India

PK Prem Meena
Associate Professor, Department of Genetics and Plant Breeding, College of Agriculture, Ummedganj, Agriculture University, Kota, Rajasthan, India

Rajesh Kumar
Assistant Professor, Department of Agronomy, College of Agriculture, Ummedganj, Agriculture University, Kota, Rajasthan, India

DL Yadav
Assistant Professor, Department of Plant Pathology, College of Agriculture, Ummedganj, Agriculture University, Kota, Rajasthan, India

Corresponding Author:
Himanshi
M. Sc. Scholar, College of Agriculture, Ummedganj, Agriculture University, Kota, Rajasthan, India

Genetic diversity analysis in M_6 generation of urdbean [*Vigna mungo* (L.) Hepper]

Himanshi, Khajan Singh, PK Prem Meena, Rajesh Kumar and DL Yadav

Abstract
The present investigation was conducted in an augmented randomized complete block design during *kharif*, 2022 with 190 mutant genotypes of urdbean varieties (Pratap Urd -1 and CO-6 treated with three doses of gamma radiation *i.e*., 200 Gy, 300 Gy and 400 Gy) including three check varieties *viz*., CO-6, Pratap Urd-1 and Kota Urd-3 and 13 component characters were studied for genetic diversity by using D^2 statistics. The genotypes were grouped into 14 distinct non-overlapping clusters, cluster VI had maximum of 40 genotypes followed by cluster III contain 33 genotypes, cluster V contain 24 genotypes, cluster I and cluster II contain 16 genotypes each while, the cluster XIII contain single genotype only. Cluster VIII had maximum intra-cluster distance. Inter-cluster distance was highest between cluster XI and XIII followed by cluster XIII and XIV, cluster IV and XIII, cluster IX and XIII, cluster II and XIII, indicating efficient breeding program can be undertaken to improve productivity and other yield attributes by selecting superior genotypes in segregating generation.

Keywords: Urdbean [*Vigna mungo* (L.) Hepper], genetic diversity, cluster, D^2 statistics

Introduction
Pulses also known as grain legumes are important source of vegetable protein. These are also important source of vitamins, complex carbohydrates, minerals and fibre. Legumes are relatively sustainable crop as these crops release less greenhouse gas emission, can sequester carbon in soil, make their own nitrogen from the atmosphere, thus reducing the application of nitrogen fertilizers. Urdbean [*Vigna mungo* (L.) Hepper] is an important grain legume with easily digestible protein and flatulence contents. Urdbean [*Vigna mungo* (L.) Hepper] is a self-pollinated, annual, short-duration, dicotyledonous legume crop with chromosome number 2 n=22 and belongs to family *fabaceae* sub-family *papilionaceae*. Urdbean grain contains about 25% protein, 56% carbohydrate, 2% fat, 4% minerals and 0.4% vitamins (vitamin A, B_1 & B_3) (Tank *et al*. (2018) [10].

India is the world's largest producer and consumer of urdbean. Madhya Pradesh, Rajasthan, Andhra Pradesh, Uttar Pradesh, Tamil Nadu, Maharashtra, Jharkhand, Gujarat, Karnataka and West Bengal are the major contributors of urdbean production in India. Urdbean production was recorded 27.8 lakh tonnes with an area of 46.3 lakh ha and productivity of 599 kg/ha in India during 2021-22. In Rajasthan the production of urdbean was recorded 1.56 lakh tonnes with area 4.25 lakh ha and productivity 366 kg/ha (Anonymous 2022-23) [1]. Major urdbean growing districts in Rajasthan are Bhilwara, Chittorgarh, Udaipur, Rajsamand, Banswara, Dungarpur, Bundi, Baran and Tonk. (Anonymous 2021-22) [2].

To meet the needs of exponential population growth, we need to improve our existing plant varieties and make them more genetically diverse, more adaptable to climate change, efficient use of inputs, high-yielding and better adaptability to a wide range of agro-ecosystems. Genetic variability in urdbean is a limiting factor due to the papilionaceous nature of the flower, which hampers its hybridization program. The major constraints in urdbean genetic improvement are lack of exploitable genetic variability, absence of suitable ideotype for different cropping systems, poor harvest index and susceptibility to biotic and abiotic stresses and non- availability of quality seeds of improved varieties (Priya *et al*. 2018) [9]. Breeding methods using hybridization have improved yield potential of urdbean crop but due to small flowers, it is tedious, burdensome, time-consuming and complicated.

Among the various breeding methods to improve crop varieties, induced mutation is one of the prominent methods of creation of genetic variation. Genetic diversity is an important parameter in any crop improvement program, which is an essential requirement for obtaining desirable segregants.

The diversity of plant genetic resources offers plant breeders the opportunity to develop new and improved cultivars that have high yield potential and are resistant to biotic and abiotic factors.

Material and Methods

The present investigation was carried out during *kharif*, 2022 at experimental field of AICRP on MULLaRP, Agricultural Research Station, Ummedganj, Agriculture University, Kota, Rajasthan, India. The experimental material consisted of 190 fixed mutant genotypes of urdbean varieties Pratap Urd-1 and CO-6 (treated with three doses of gamma radiation *i.e.*, 200 Gy, 300 Gy and 400 Gy) obtained from AICRP on MULLaRP, Agricultural Research Station, Ummedganj, Kota including three check varieties *viz.*, CO-6, Pratap Urd-1 and Kota Urd-3. The experiment was laid out in an augmented randomized complete block design (Federer, 1956) [3]. The material was sown in 10 blocks. Each block had 5 meter long two-rowed 22 plots placed 30 cm apart. Thus 19 genotypes and 3 checks were sown in each block. The observations were recorded on five randomly selected plants for all characters from each genotype in each block, except for days to 50% flowering and days to maturity which recorded on whole plot basis.

Result and Discussion

Analysis of variance (Table-1) revealed that mean square due to genotypes were significant for all the traits except days to maturity indicating a good amount of variability in the experimental material. The mean square due to checks were also found significant for all the traits except days to 50 per cent flowering indicating that checks were also diverse. Genetic diversity analysis was carried out by calculating D^2 values from means of 190 genotypes of urdbean for 13 characters. The genotypes were grouped into 14 clusters (Table-2). Earlier workers have also reported existence of substantial genetic divergence by Panigrahi *et al.* (2014) [8], Gowsalya *et al.* (2017) [5], Goswami *et al.* (2022) [4] and Kuralarasan *et al.* (2018) [7]. Among the 14 clusters, cluster VI contains maximum number of 40 genotypes followed by cluster III contain 33 genotypes, cluster V contain 24 genotypes, cluster I and cluster II contain 16 genotypes each.

Cluster VIII comprises 14 genotypes, cluster VII contains 13 genotypes, cluster IV contains 9 genotypes, cluster XIV contain 8 genotypes, cluster IX and cluster X comprises 7 genotypes each, cluster XII contain 3 genotypes, cluster XI contain 2 genotypes while, the cluster XIII contain single genotype only. The dendrogram depicting the clustering pattern was depicted in figure 1. The discriminant of genotypes in so many discrete clusters, suggested presence of high degree of genetic diversity in the material evaluated.

The intra and inter cluster D^2 values are presented in the table-3. Inter-cluster distances were greater than intra-cluster distances which shows considerable amount of genetic diversity existed among the genotypes. Similar reports were given by Panigrahi *et al.* (2014) [8] and Gowsalya *et al.* (2017) [5]. Out of 14 clusters, cluster VIII had maximum intra-cluster distance followed by cluster III, IV and IX. This implies that these groups were diversed. The cluster XIII showed zero intra-cluster distance due to monogenotypic nature. High intra-cluster genetic distance in cluster VIII was because of heterogeneous composition of that cluster. Inter-cluster distances ranged from 3.26 to 42.76. Maximum inter-cluster genetic distance was observed between cluster XI and XIII (42.76) followed by cluster XIII and XIV (32.54), cluster IV and XIII (31.48) and cluster IX and XIII (30.09). Maximum inter-cluster distances, indicating a high degree of genetic diversity. As a result, genotypes from the most diversified cluster could be employed as parents in hybridization programmes to create high yielding cultivars. The least inter-cluster distance was found between cluster I and II (3.26) suggested that the genetic constitution of the genotype in one cluster were in close proximity with the genotype in the other cluster of the pair. Hence, genotypes from these clusters may not be useful. This result is supported by findings of Kumar *et al.* (2014) [6] and Veni *et al.* (2016) [11] and Kuralarasan *et al.* (2018) [7].

Cluster XIII includes genotypes with higher number of seeds per pod, harvest index, protein content and high seed yield per plant, a greater number of clusters per plant and more number of pods per plant. Cluster XI for dwarf plant, cluster XII for a greater number of branches per plant, Cluster VIII include with tall plants, more number of branches per plant, high number of clusters per plant, number of pods per plant, high biological yield per plant and seed yield per plant. Cluster VII is desirable for early flowering, long pod and high 100 seed index.

The cluster means for the 13 characters are given in table-4. The lowest cluster mean value was exhibited for days to 50% flowering cluster VII and for days to maturity was recorded by cluster I. The highest mean for number of clusters per plant, number of pods per plant and biological yield per plant was observed in cluster VIII, highest cluster mean for pod length was observed in cluster II and for number of seeds per pod and seed yield per plant was observed in cluster XIII. Hence, the genotypes in this cluster were high yielding.

Table 1: Analysis of variance (ANOVA) for different traits in Urdbean

Source of variation	D.F.	Mean Sum of Square												
		Days to 50 percent flowering	Days to maturity	Plant height (cm)	Number of branches per plant	Number of clusters per plant	Number of pods per plant	Pod length (cm)	Number of seeds per pod	Biological yield per plant (g)	100 – seed weight (g)	Harvest index (%)	Protein content (%)	Seed yield per plant (g)
Blocks (b-1)	9	1.11	0.30	7.29	0.41	1.40	2.61	0.178**	0.26	2.38	0.07	60.15	0.11	0.15
Entries (c+g)-1	192	1.87**	3.68**	32.82**	2.68**	3.99**	23.74**	0.127**	0.70**	4.39**	0.23**	87.39**	1.06**	1.02**
Checks (c-1)	2	0.63	101.03**	51.90**	4.94**	25.44**	184.81**	0.47**	2.35**	12.17**	0.94**	1611.1**	2.29**	9.06**
Genotypes (g-1)	189	1.80**	2.35	32.60**	2.29**	3.78**	22.14**	0.12**	0.69**	4.33**	0.23**	71.55**	1.00**	0.95**
Genotypes v/s Checks	1	16.44**	58.84**	36.37**	71.87**	0.06	3.50	0.02	0.46	0.31	0.02	33.46	9.10**	0.02
Error (b-1) (c-1)	18	0.52	1.25	7.53	0.21	0.68	1.11	0.06	0.17	1.21	0.08	31.87	0.28	0.06

** significant at 1% significance level

Table 2: Cluster composition of 190 mutant genotypes of urdbean

Clusters	Number of genotypes	Genotypes
Cluster I	16	KPU 100-2, KPU 100-4, KPU 100-13 A, KPU 200-4, KPU 200-6, KPU 300-76 A, KPU 300-75 C, KPU 300-76B, KPU 300-8B, KPU 400-45 B1, KPU 400-47A, KPU 400-53B, KPU 400-56A, KPU 400-67A2, KPU 400-76C2 and KPU 400-77 A1
Cluster II	16	KPU100-8, KPU 200-8A, KPU 200-8B, KPU 200-9A, KPU 200-9B, KPU 200-10A, KPU 200-10B, KPU 200-10C, KPU 200-11, KPU 200-25B, KPU 200-35A, KPU 300-36B, KPU 300-51C, KPU 300-104D, KPU 400-46A2 and KPU 400-66C
Cluster III	33	KPU 100-13B, KPU200-1, KPU 200-2, KPU 200-16C, KPU 200-21A, KPU 200-22, KPU 200-23, KPU 200-24, KPU 200-28A, KPU 200-29A, KPU 200-29B, KPU 200-30A, KPU 200-32B, KPU 200-33A, KPU 200-35B, KPU 200-43, KPU 200-49, KPU 200-48, KPU 400-12, KPU 400-102, KPU 300-100A, KPU 300-104E, KPU 300-116, KPU 300-123B, KPU 300-124A, KPU 300-124, KPU 400-41B, KPU 400-43B1, KPU 400-45 B2, KPU 400-46 A1, KPU 400-46A3, KPU 400-86E and KPU 400-86D
Cluster IV	9	KPU 200-8C, KPU 200-21C, KPU 200-28B, KPU 400-17, KPU 300-41D, KPU 400-38A, KPU 400-43B2, KPU 400-64A and CO-6
Cluster V	24	KPU 200-12A, KPU 200-12B, KPU 200-12C, KPU 200-13 A, KPU 200-13B, KPU 200-16A, KPU 200-16B, KPU 200-20A, KPU 200-20B, KPU 200-20C, KPU 200-21B, KPU 200-40, KPU 300-78A, KPU 300-2 B, KPU 300-37, KPU 300-40, KPU 300-45D, KPU 300-18 C, KPU 300-120B, KPU 400-34, KPU 400-86C1, KPU 400-86C2, KPU 500-68C and Kota Urd-3
Cluster VI	40	KPU 200-25A, KPU 200-33B, KPU 200-50, KPU 200-51A, KPU 200-51B, KPU 200-54, KPU 300-1, KPU 300-2A, KPU 300-3, KPU 300-6, KPU 400-91, KPU 200-69, KPU 300-1B, KPU 300-47C, KPU 300-46C, KPU 300-63, KPU 300-114A, KPU 300-114B1, KPU 300-114 B2, KPU 300-118B, KPU 300-119, KPU 300-120A, KPU 300-123A, KPU 300-124B1, KPU 300-124B2, KPU 400-45B3, KPU 400-45B4, KPU 400-46C, KPU 400-55A2, KPU 400-64B, KPU 400-68, KPU 400-76B2, KPU 400-77A2, KPU400-78A, KPU 400-78B, KPU 400-82 A, KPU 400-88, KPU 400-100A,KPU 400-26A and Pratap Urd -1
Cluster VII	13	KPU 200-30B, KPU 200-32A, KPU 200-39, KPU 200-41, KPU 200-45, KPU 400-47, KPU 200-52, RBU-38, KPU 300-4A, KPU 300-7B, KPU 300-8A, KPU 300-9A and KPU 300-76C
Cluster VIII	14	KPU 300-5A, KPU 300-7A, KPU 200-71, KPU 300-1C, KPU 300-2A, KPU 300-2C, KPU 300-4B, KPU 300-5B, KPU 300-9B, KPU 300-18A, KPU 300-26, KPU 400-76C1, KPU 400-86 and KPU 400-86B
Cluster IX	7	KPU 200-72, KPU 400-53A, KPU 400-55A1, KPU 400-67A1, KPU 400-73, KPU 400-76B1 and KPU 400-82B
Cluster X	7	KPU 300-41A, KPU 300-50, KPU 300-79, KPU 300-99A, KPU 300-99 C, KPU 300-72 A and KPU 300-72B
Cluster XI	2	KPU 300-114C and KPU 400-45 A
Cluster XII	3	KPU 300-115 A, KPU 300-115C1 and KPU 300-115 C2
Cluster XIII	1	KPU 400-83
Cluster XIV	8	KPU 500-39C, KPU 500-42, KPU 500-43A, KPU 500-43 C, KPU 500-65, KPU 500-66, KPU 500-68A and KPU 500-68B

Table 3: Inter and Intra- cluster distance among seed yield and its attributing traits

Cluster	Cluster I	Cluster II	Cluster III	Cluster IV	Cluster V	Cluster VI	Cluster VII	Cluster VIII	Cluster IX	Cluster X	Cluster XI	Cluster XII	Cluster XIII	Cluster XIV
Cluster I	8.77	3.26	4.31	7.07	8.32	6.33	7.60	13.19	6.27	10.43	17.00	7.53	26.91	8.55
Cluster II		8.11	3.53	5.36	10.38	7.93	9.64	14.94	5.98	12.92	14.57	7.53	29.32	6.75
Cluster III			8.92	4.68	9.48	5.94	9.14	16.06	8.51	12.45	15.32	9.34	28.17	5.90
Cluster IV				8.83	13.01	9.45	11.80	17.19	8.55	15.84	12.54	9.92	31.48	6.78
Cluster V					8.09	5.16	4.08	11.74	12.18	4.43	24.26	11.92	19.15	14.46
Cluster VI						8.64	5.10	13.85	10.81	7.88	20.62	10.41	22.67	10.67
Cluster VII							6.43	9.71	10.33	5.55	23.24	10.06	20.90	14.31
Cluster VIII								10.77	11.92	11.23	27.20	11.23	23.10	20.90
Cluster IX									8.78	14.02	15.95	5.52	30.09	11.31
Cluster X										5.58	27.01	13.14	17.21	17.35
Cluster XI											5.89	17.58	42.76	11.86
Cluster XII												5.95	28.94	12.91
Cluster XIII													0.00	32.54
Cluster XIV														6.73

Table 4: Clusters mean of seed yield and its attributing traits

Cluster	Days to 50 percent flowering	Days to maturity	Plant height (cm)	Number of branches per plant	Number of clusters per plant	Number of pods per plant	Pod length (cm)	Number of seeds per pod	Biological yield per plant (g)	100 – seed weight (g)	Harvest index (%)	Protein content (%)	Seed yield per plant (g)
Cluster I	42.58	71.44	31.14	7.90	4.93	10.32	4.07	4.12	6.70	4.12	30.61	23.68	1.87
Cluster II	42.00	71.65	30.09	7.42	4.59	9.92	4.64	4.61	5.61	3.81	28.53	22.16	1.51
Cluster III	41.91	71.47	27.51	7.48	5.09	9.99	4.18	4.05	5.03	3.94	30.67	22.40	1.59
Cluster IV	40.97	71.47	25.61	6.81	6.02	11.07	3.96	4.38	6.19	3.36	27.10	22.95	1.68
Cluster V	40.98	72.24	32.37	6.98	5.60	13.11	4.04	4.56	5.99	4.20	37.93	22.70	2.21
Cluster VI	41.30	72.53	28.55	9.10	5.46	11.87	4.06	4.72	5.84	3.93	35.60	23.21	2.06
Cluster VII	39.54	71.70	32.17	9.17	6.40	14.14	4.36	5.17	7.15	4.34	35.72	23.31	3.01
Cluster VIII	41.02	72.50	38.27	9.21	8.09	20.19	4.12	4.72	8.79	4.13	32.46	24.24	3.27
Cluster IX	40.71	72.95	33.53	8.88	6.39	12.19	4.03	4.01	5.89	3.26	26.19	24.02	1.48
Cluster X	42.14	71.76	34.46	8.80	6.67	12.93	3.89	5.62	8.24	4.15	39.58	22.64	2.69
Cluster XI	41.00	74.07	23.13	7.50	2.82	5.55	3.11	2.44	4.60	2.85	17.36	22.58	0.65
Cluster XII	42.67	74.90	32.78	10.92	6.27	13.85	4.00	3.70	7.51	4.08	27.51	20.83	1.82
Cluster XIII	41.33	75.23	36.88	7.49	6.78	18.01	3.91	5.94	7.95	3.58	54.98	24.96	5.34
Cluster XIV	42.46	71.48	25.49	7.97	2.48	6.92	3.75	4.76	2.89	2.95	28.07	23.25	1.12

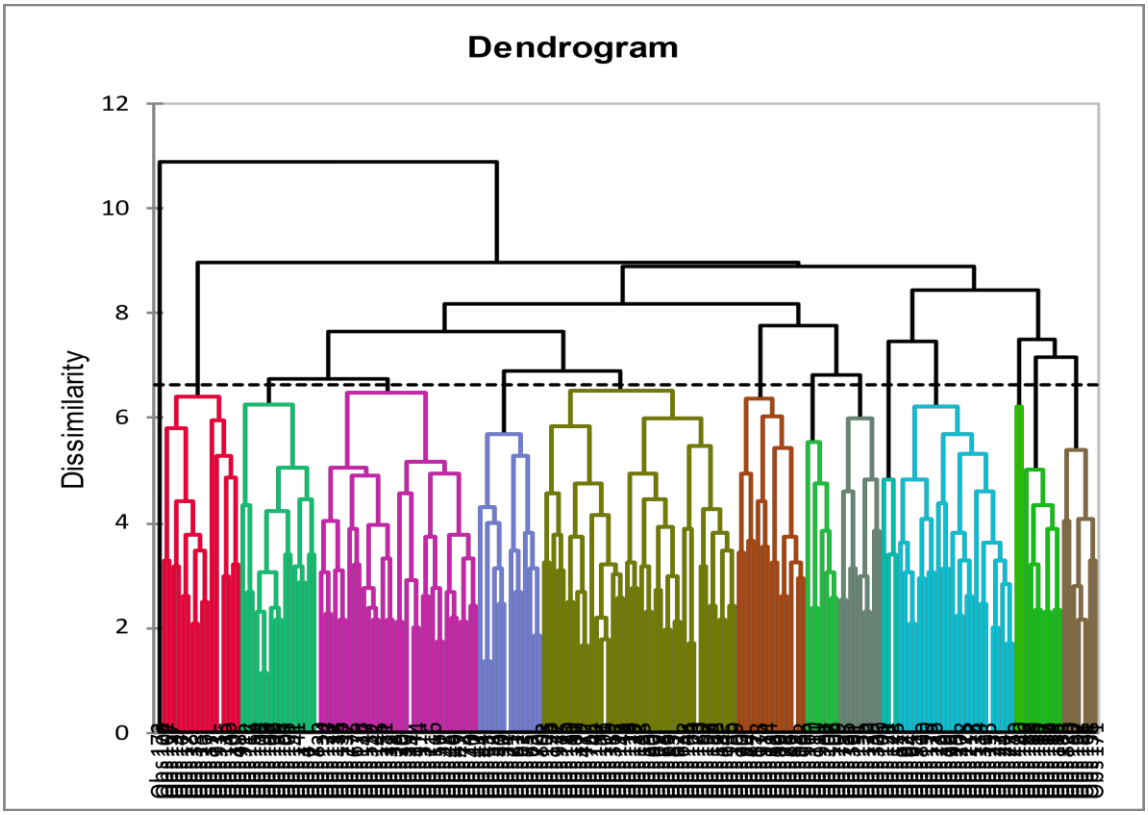

Fig 1: Dendrogram depicting the clustering pattern of 190 mutant genotypes of urdbean

Conclusion

Genotypes falls in group XI and XIII and cluster XIII and XIV were highly diverse from each-other and can be used has divergent parents for hybridization. Cluster XIII consist genotypes with higher number of seeds per pod, seed yield per plant, protein content high harvest index, while cluster VIII include number of clusters per plant, number of pods per plant, biological yield per plant. Genotypes KPU 300-1C, KPU 300-4B, KPU 300-18A and KPU 300-26 were found promising for number of branches per plant, number of clusters per plant, number of pods per plant, biological yield per plant and seed yield per plant in cluster VIII, while KPU 300-4A genotype in cluster VII and KPU 400-83 in cluster XIII were found suitable for early flowering, number of seeds per plant and seed yield per plant.

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