



Research article

Phenotypic diversity in mutagenized population of urdbean (*Vigna mungo* (L.) Hepper)Sonu Goyal^a, Mohammad Rafiq Wani^{b,*}, Aamir Raina^{a,c}, Rafiul Amin Laskar^d, Samiullah Khan^a^a Mutation Breeding Laboratory, Department of Botany, Aligarh Muslim University, Aligarh-202 002, Uttar Pradesh, India^b Department of Botany, Abdul Ahad Azad Memorial Degree College Bemina, Cluster University Srinagar, Jammu and Kashmir, 190 018, India^c Botany Section, Women's College, Aligarh Muslim University, Aligarh-202 002, Uttar Pradesh, India^d Department of Botany, Bahona College, Jorhat-785 101, Assam, India

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ABSTRACT

The present study was conducted to assess the extent of induced genetic variability and to determine the inheritance pattern of various yield contributing phenotypic traits in M_2 and M_3 generations of urdbean following mutagenesis with single and combination treatments of gamma rays and ethyl methanesulphonate (EMS). The mean number of seeds per pod and 100-seed weight increased in all the mutagenic treatments in both the varieties with few exceptions in M_2 generation. Mean pod length although increased considerably, however it did not differ significantly in most of the mutagenic treatments. In M_3 generation, 0.2% EMS and 300 Gy γ rays+0.2% EMS treatments induced maximum increase in mean pod length, seeds per pod and 100-seed weight in both the varieties. Genetic parameters showed manifold increase in most of the mutagenic treatments and varied from trait to trait. Increased genetic variability for economically important traits in the selected mutant lines has successfully contributed in diversifying the accessible genetic base which could be exploited for subsequent improvement of urdbean through phenotypic selection.

1. Introduction

Crop plants are genetically very diverse groups grown in temperate, subtropical and tropical regions and have been recognized for their human health benefits by containing high contents of non-nutritive, nutritive and bioactive compounds such as flavonoids, phenolics, sugars, essential oils, minerals etc (Zia-ul-Haq et al., 2014; Oktem, 2019; Unlukara, 2019). Urdbean (*Vigna mungo* (L.) Hepper) is an important rainfed short duration legume crop of India and is considered as an excellent source of high-quality dietary protein with good digestibility. It contributes a major portion of lysine in vegetarian diet of most of the Asian population (Gill et al., 2017). However, its productivity is stumpy due to low yield potential and narrow genetic base of the existing cultivars. In these circumstances, induced mutagenesis may be employed for developing high yielding genotypes by creating heritable variation in yield and yield attributing traits (Baisakh et al., 2011; Wani, 2017, 2021, Amin et al., 2019; Goyal et al., 2019; Kumar et al., 2019; Raina and Khan, 2020; Raina et al., 2020).

Induced mutations are exploited to generate extensive genetic variability for developing mutants with desirable agronomical traits (Laskar et al., 2019). Since inheritance of quantitative traits is controlled by

interaction of different genes with additive effects on phenotypic variability, the estimates of genetic parameters like genotypic coefficient of variation, heritability and genetic advance in segregating mutant populations are indispensable to foresee the scope of improvement vis-à-vis the target traits through selection (Laskar et al., 2018). Lately, induced mutations were found to be effective in creating genetic variability for different quantitative traits in various pulse crops like mungbean (Singh, 2009), chickpea (Raina et al., 2019), lentil (Laskar et al., 2018), field pea (Singh et al., 2015), cowpea (Raina et al., 2018), black gram (Goyal et al., 2020), fenugreek (Hassan et al., 2018), black cumin (Tantray et al., 2017), pigeon pea (Etther et al., 2019) and faba bean (Khurshed et al., 2018).

The FAO/IAEA Mutant Variety Database (<https://mvd.iaea.org> accessed on 24 January, 2021) records show that out of total 466 released mutant varieties of legumes, only 9 have been released in urdbean till date, indicating that the crop is less exploited for mutation breeding. Therefore, sustained efforts are needed to develop reproducible mutation protocols for creating novel genes controlling economically important traits, especially in India where the scope for agricultural intensification is very high (Laskar and Khan, 2017).

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To augment the existing genetic variability for desirable traits, the present investigation was carried out to induce additional genetic diversity among different interrelated polygenic traits viz., pod length, number of seeds per pod and 100 seed weight in M_2 and M_3 generations for developing luxuriant, high yielding, disease resistant and stabilized mutants of urdbean.

2. Materials and methods

Urdbean varieties T-9 and Pant U-30 were developed by local selection from Bareilly, Uttar Pradesh, India and pedigree method of selection from the cross between UPU-1 X UPU-2, respectively. Healthy and viable seeds of varieties T-9 and Pant U-30 were obtained from Govind Ballabh Pant University of Agriculture and Technology, Pantnagar, Uttaranchal, India. Dry seeds with 12% moisture content were directly irradiated with 100 Gy, 200 Gy, 300 Gy and 400 Gy of gamma (γ) rays from ^{60}Co (cobalt-60) source at National Botanical Research Institute (NBRI), Lucknow, India. The percentage of moisture content was determined following the guidelines of International Seed Testing Association (ISTA, 1985) based on the difference between fresh and dry weights of the seeds. For chemical treatments, fresh aqueous 1% (v/v) stock solution of EMS, manufactured by Sissco Research Laboratories Pvt. Ltd., Mumbai, India, were prepared in phosphate buffer of pH 7.0. Seeds were pre-soaked in double distilled water for 9 hours prior to treatment with 0.1%, 0.2%, 0.3% and 0.4% EMS prepared from stock solution for 6 hours under standard conditions at Mutation Breeding Laboratory, Aligarh Muslim University (A.M.U.), Aligarh. Following chemical treatments, the seeds were thoroughly washed under running tap water to remove the residual mutagen adhered on seed coat. For combination treatments, 200 Gy γ rays+0.2% EMS, 300 Gy γ rays+0.2% EMS, 200 Gy γ rays+0.3% EMS

and 300 Gy γ rays+0.3% EMS were prepared by directly treating the gamma irradiated seeds with EMS concentrations.

Three replications of 100 seeds each were sown for every treatment and controls in randomized complete block design (RCBD) to raise M_1 generation. The experiment was carried out at the Agriculture Farm, A.M.U., Aligarh, India. The spacing was maintained at 0.30 m (seed to seed in a row) and 0.60 m (between the rows) inside each block. Recommended agronomic practices were employed for field preparation, seed sowing and afterward management of the crop in all the generations. All the fertile M_1 plants were harvested individually and twenty-five healthy seeds from each harvested plant were stored and sown in plant progeny row basis to grow M_2 generation. For raising M_3 generation, 10 such M_2 progenies from the treatments of 200 and 300 Gy γ rays, 0.2 and 0.3% EMS and 200 Gy γ rays+0.2% EMS and 300 Gy γ rays+0.2% EMS treatments were selected which showed significant positive shift in mean values. For statistical analysis, 30 best performing plants were considered from each treatment. Data collected for quantitative phenotypic traits such as pod length, number of seeds per pod and 100-seed weight in M_2 and M_3 generations were subjected to one-way analysis of variance to assess the extent of induced phenotypic and genotypic variation (Singh and Chaudhary, 1985). The significance of difference between the means of treated and control populations was tested by using critical difference (CD) estimated from error mean square and tabulated 'T' value at 5% level of significance.

3. Results and discussion

Genetic architecture of grain yield can be better resolved through component traits than yield *per se*, as yield is the end product of multiple interactions between various component traits (Laskar and Khan, 2017).

Table 1. Effects of gamma (γ) rays, EMS and their combination treatments on seed germination and fertility in varieties T9 and Pant U-30 of urdbean in M_1 generation.

Treatments	Seed germination (%)		Fertility (%)	
	Actual	%age inhibition	Actual	%age inhibition
Var. T-9				
Control	89.00	-	99.63	-
100 Gy γ rays	82.67	7.12	96.77	2.87
200 Gy γ rays	79.33	10.86	93.28	6.38
300 Gy γ rays	76.67	13.86	90.43	9.23
400 Gy γ rays	73.00	17.98	86.76	12.92
0.1% EMS	81.33	8.61	95.49	4.15
0.2% EMS	77.33	13.11	91.38	8.28
0.3% EMS	75.00	15.73	88.89	10.78
0.4% EMS	71.67	19.48	85.12	14.57
200 Gy γ rays +0.2% EMS	78.33	11.99	92.34	7.32
300 Gy γ rays +0.2% EMS	74.00	16.85	87.84	11.84
200 Gy γ rays +0.3% EMS	70.67	20.60	83.96	15.73
300 Gy γ rays +0.3% EMS	69.33	22.10	82.69	17.00
Var. Pant U-30				
Control	86.67	-	98.85	-
100 Gy γ rays	78.67	9.23	90.68	8.27
200 Gy γ rays	75.00	13.46	86.67	12.33
300 Gy γ rays	72.33	16.54	83.41	15.62
400 Gy γ rays	68.67	20.77	79.13	19.95
0.1% EMS	80.00	7.70	92.50	6.42
0.2% EMS	77.00	11.16	88.74	10.22
0.3% EMS	74.00	14.62	85.14	13.87
0.4% EMS	70.00	19.23	80.48	18.59
200 Gy γ rays +0.2% EMS	75.67	12.70	87.22	11.76
300 Gy γ rays +0.2% EMS	71.67	17.31	82.79	16.25
200 Gy γ rays +0.3% EMS	68.00	21.54	78.43	20.66
300 Gy γ rays +0.3% EMS	66.33	23.46	76.38	22.73

Table 2. Estimates of mean values (\bar{X}), shift in \bar{X} and genetic parameters for pod length (cm) in M_2 generation of urdbean var. T-9*.

Treatment	Mean \pm S.E.	Shift in \bar{X}	PCV (%)	GCV (%)	h^2 (%)	GA (% of \bar{X})
Control	4.25 \pm 0.03	-	4.34	2.10	26.67	2.82
100 Gy γ rays	4.36 \pm 0.05	+0.11	9.17	5.04	31.25	7.57
200 Gy γ rays	4.60 \pm 0.09	+0.35	10.22	6.00	36.36	9.78
300 Gy γ rays	4.72 \pm 0.06	+0.47	11.65	8.82	46.67	14.19
400 Gy γ rays	4.02 \pm 0.07	-0.23	9.86	4.22	20.00	4.97
CD (p = 0.05)		0.48				
0.1% EMS	4.29 \pm 0.10	+0.04	9.09	4.66	26.67	6.29
0.2% EMS	4.87 \pm 0.07	+0.62	8.00	5.15	42.85	8.62
0.3% EMS	4.80 \pm 0.06	+0.55	9.79	6.04	36.36	9.37
0.4% EMS	4.05 \pm 0.08	-0.20	5.18	2.72	25.00	3.46
CD (p = 0.05)		0.57				
200 Gy γ rays +0.2% EMS	4.21 \pm 0.10	-0.04	7.60	3.56	20.00	4.04
300 Gy γ rays +0.2% EMS	4.52 \pm 0.11	+0.27	9.73	5.68	36.84	9.29
200 Gy γ rays +0.3% EMS	4.62 \pm 0.08	+0.37	8.58	5.10	33.33	7.36
300 Gy γ rays +0.3% EMS	3.99 \pm 0.06	-0.26	6.26	3.01	23.33	3.76
CD (p = 0.05)		0.43				

*SE = Standard error; PCV = Phenotypic coefficient of variation; GCV = Genotypic coefficient of variation; h^2 = Heritability; GA = Genetic advance; CD = Critical difference.

In the present investigation, the observations on different quantitative traits revealed a wide extent of genetic variability induced through individual and combination treatments of gamma rays and EMS. Effects of mutagens on seed germination and fertile plants in M_1 generation showed differential sensitivity of both the varieties towards different mutagenic treatments (Table 1). This revealed that rate of induced mutation(s) is dependent on the specific interaction between genotype and mutagen dose. In M_2 generation, mean values for quantitative traits shifted towards positive side in most of the mutagenic treatments. The variation in the mean values due to small and cumulative effects of multiple genes governing quantitative traits are discussed below:

3.1. Pod length (cm)

In M_2 generation, the results revealed that most of the mutagenic treatments were not capable of inducing significant differences in mean pod length in both the varieties. The mean pod length was recorded as 4.25 and 4.40 cms in the control populations of the varieties T-9 and Pant U-30, respectively. The maximum increase in mean pod length was recorded in 0.2% EMS treatment in the varieties T-9 (4.87) and Pant U-30 (5.08) (Tables 2 and 3). The estimated genotypic coefficient of variation

(8.82 and 8.46%), heritability (46.67 and 41.86%) and genetic advance (14.19 and 13.97%) was maximum in 300 Gy γ rays and 0.2% EMS treatments in the varieties T-9 and Pant U-30, respectively (Tables 2 and 3). In M_3 generation, a significant increase in mean pod length was recorded in 0.2% EMS treatment in the varieties T-9 (4.92) and Pant U-30 (5.11). The estimated genotypic coefficient of variation was maximum in 0.2% EMS and 200 Gy γ ray treatments in the varieties T-9 (6.56%) and Pant U-30 (7.28%), respectively. The highest heritability was recorded in 200 Gy γ rays and 0.3% EMS treatments in the varieties T-9 (34.78%) and Pant U-30 (41.93%) respectively. The maximum genetic advance (9.43 and 12.20%) was recorded in 0.3% EMS treatment in the varieties T-9 and Pant U-30, respectively (Table 4).

The increased mean pod length can have direct impact on significant improvement of other traits such as seeds per pod, 100-seed weight and seed yield as longer pods can accommodate extra and bold seeds. Increased pod length can accommodate more number of seeds per pod, thus leading to enhanced yield potential (Wani et al., 2011). Moreover, it was also observed that the increase in mean pod length was associated with low genetic variability in all the treatments, suggesting a limited scope of its further improvement in succeeding generations.

Table 3. Estimates of mean values (\bar{X}), shift in \bar{X} and genetic parameters for pod length (cm) in M_2 generation of urdbean var. Pant U-30.

Treatment	Mean \pm S.E.	Shift in \bar{X}	PCV (%)	GCV (%)	h^2 (%)	GA (% of \bar{X})
Control	4.40 \pm 0.05	-	4.00	1.96	26.67	2.73
100 Gy γ rays	4.42 \pm 0.06	+0.02	9.95	4.98	26.31	6.78
200 Gy γ rays	4.76 \pm 0.06	+0.36	10.20	3.95	17.39	4.41
300 Gy γ rays	4.85 \pm 0.07	+0.40	8.45	5.06	35.29	7.83
400 Gy γ rays	4.11 \pm 0.08	-0.29	9.24	4.14	21.43	5.11
CD (p = 0.05)		0.39				
0.1% EMS	4.45 \pm 0.05	+0.05	10.25	4.22	19.05	5.17
0.2% EMS	5.08 \pm 0.09	+0.68	12.99	8.46	41.86	13.97
0.3% EMS	4.98 \pm 0.07	+0.58	11.25	7.03	38.71	11.24
0.4% EMS	4.20 \pm 0.10	-0.20	7.38	3.09	16.67	3.09
CD (p = 0.05)		0.59				
200 Gy γ rays +0.2% EMS	4.44 \pm 0.05	+0.04	7.88	4.50	33.33	6.75
300 Gy γ rays +0.2% EMS	4.73 \pm 0.06	+0.33	11.20	6.98	39.28	11.41
200 Gy γ rays +0.3% EMS	4.68 \pm 0.08	+0.28	9.26	4.88	27.78	6.62
300 Gy γ rays +0.3% EMS	4.00 \pm 0.07	-0.40	7.25	3.25	25.00	4.75
CD (p = 0.05)		0.29				

Table 4. Estimates of mean values (\bar{X}), shift in \bar{X} and genetic parameters for pod length (cm) in M_3 generation of urdbean.

Treatment	Mean \pm S.E.	Shift in \bar{X}	PCV (%)	GCV (%)	h^2 (%)	GA (% of \bar{X})
Var. T-9						
Control	4.30 \pm 0.02	-	6.52	3.25	25.64	4.19
200 Gy γ rays	4.71 \pm 0.06	+0.41	10.20	6.12	34.78	9.13
300 Gy γ rays	4.76 \pm 0.09	+0.46	11.76	5.88	25.81	7.77
CD (p = 0.05)		0.52				
0.2% EMS	4.92 \pm 0.07	+0.62	11.19	6.32	30.00	8.73
0.3% EMS	4.88 \pm 0.04	+0.58	11.47	6.56	31.25	9.43
CD (p = 0.05)		0.38				
200 Gy γ rays +0.2% EMS	4.59 \pm 0.10	+0.29	8.06	3.66	20.71	4.14
300 Gy γ rays +0.2% EMS	4.62 \pm 0.05	+0.32	9.31	5.19	33.33	7.79
CD (p = 0.05)		0.49				
Var. Pant U-30						
Control	4.42 \pm 0.05	-	7.47	3.47	18.18	3.62
200 Gy γ rays	4.81 \pm 0.10	+0.39	12.06	7.28	35.29	11.02
300 Gy γ rays	4.90 \pm 0.08	+0.48	10.61	6.53	37.04	10.41
CD (p = 0.05)		0.49				
0.2% EMS	5.11 \pm 0.07	+0.69	9.25	5.22	31.82	7.44
0.3% EMS	5.00 \pm 0.04	+0.58	11.20	7.20	41.93	12.20
CD (p = 0.05)		0.23				
200 Gy γ rays +0.2% EMS	4.69 \pm 0.09	+0.27	8.95	4.45	22.22	5.12
300 Gy γ rays +0.2% EMS	4.76 \pm 0.06	+0.34	9.45	4.01	18.00	4.41
CD (p = 0.05)		0.44				

3.2. Number of seeds per pod

Data recorded on number of seeds per pod show that the shift in mean values was bi-directional, being more towards the positive direction in M_2 generation (Tables 5 and 6). Mean number of seeds per pod increased slightly at lower and moderate mutagenic doses, however a negative shift was noticed with higher treatments in M_2 generation. Khan (1985) attributed this negative shift to the high degree of seed sterility. The mean number of seeds per pod was recorded as 6.13 and 6.25 in the control populations of the varieties T-9 and Pant U-30, respectively. The maximum increase in number of seeds per pod was recorded in 300 Gy γ rays+0.2% EMS and 200 Gy γ ray treatments in the varieties T-9 (7.53) and Pant U-30 (7.80), respectively (Tables 5 and 6). The estimated genotypic coefficient of variation (11.30 and 11.25%), heritability (52.89 and 54.90%) and genetic advance (21.18 and 21.56%) was maximum in the varieties T-9 and Pant U-30, respectively (Tables 5 and 6). The results revealed a significant increase in all the

mutagenic treatments in M_3 generation. The maximum increase in mean pod length was recorded in 300 Gy γ rays +0.2% EMS treatments in the varieties T-9 (7.82) and Pant U-30 (8.23), respectively. In M_3 generation, the highest genotypic coefficient of variation (18.87 and 19.35%), heritability (85.96 and 87.95%) and genetic advance (45.50 and 47.57%) were noticed with 200 Gy γ rays and 0.2% EMS treatments in the var. T-9 and Pant U-30, respectively (Table 7). Giri et al. (2010) in pigeon pea and Khan et al. (2005) in chickpea recorded a non-significant difference in mean number of seeds per pod after mutagenic treatments.

3.3. 100-Seed weight (g)

Data recorded on mean 100-seed weight showed a slight improvement over the controls at moderate and lower doses of gamma rays and EMS in M_2 and M_3 generations. The number of seeds per pod was recorded as 3.00 and 3.01 in the control populations of the varieties T-9 and Pant U-

Table 5. Estimates of mean values (\bar{X}), shift in \bar{X} and genetic parameters for number of seeds per pod in M_2 generation of urdbean var. T-9.

Treatment	Mean \pm S.E.	Shift in \bar{X}	PCV (%)	GCV (%)	h^2 (%)	GA (% of \bar{X})
Control	6.13 \pm 0.14	-	6.85	2.25	10.55	1.79
100 Gy γ rays	5.96 \pm 0.13	-0.17	11.74	5.92	25.00	7.55
200 Gy γ rays	7.13 \pm 0.16	+1.00	15.45	11.30	52.89	21.18
300 Gy γ rays	6.92 \pm 0.19	+0.79	8.24	5.75	46.87	9.97
400 Gy γ rays	5.66 \pm 0.19	-0.47	10.43	5.20	22.86	6.00
CD (p = 0.05)		0.71				
0.1% EMS	5.77 \pm 0.17	-0.36	10.24	6.00	34.28	9.18
0.2% EMS	7.11 \pm 0.13	+0.98	14.17	10.30	50.98	18.51
0.3% EMS	6.50 \pm 0.15	+0.37	6.90	4.40	40.00	7.23
0.4% EMS	5.03 \pm 0.25	-1.10	9.24	4.20	19.05	4.57
CD (p = 0.05)		0.34				
200 Gy γ rays +0.2% EMS	7.28 \pm 0.18	+1.15	9.22	6.40	46.67	11.13
300 Gy γ rays +0.2% EMS	7.53 \pm 0.19	+1.40	12.26	8.53	48.23	15.40
200 Gy γ rays +0.3% EMS	6.50 \pm 0.12	+0.37	7.35	4.12	30.43	5.85
300 Gy γ rays +0.3% EMS	5.06 \pm 0.27	-1.07	9.09	4.95	28.57	6.71
CD (p = 0.05)		0.57				

Table 6. Estimates of mean values (\bar{X}), shift in \bar{X} and genetic parameters for number of seeds per pod in M_2 generation of urdbean var. Pant U-30.

Treatment	Mean \pm S.E.	Shift in \bar{X}	PCV (%)	GCV (%)	h^2 (%)	GA (% of \bar{X})
Control	6.25 \pm 0.19	-	7.25	2.35	10.00	1.92
100 Gy γ rays	6.54 \pm 0.19	+0.29	13.02	7.74	36.11	12.38
200 Gy γ rays	7.80 \pm 0.17	+1.55	9.65	6.46	44.64	11.15
300 Gy γ rays	7.06 \pm 0.19	+0.81	12.09	8.32	48.61	15.29
400 Gy γ rays	5.68 \pm 0.16	-0.57	11.25	6.35	31.71	9.15
CD (p = 0.05)		0.55				
0.1% EMS	6.03 \pm 0.24	-0.22	14.20	9.68	45.94	16.91
0.2% EMS	7.25 \pm 0.14	+1.00	14.56	10.15	48.18	18.34
0.3% EMS	6.68 \pm 0.16	+0.43	15.20	11.25	54.90	21.56
0.4% EMS	5.10 \pm 0.25	-1.15	13.82	7.43	28.57	10.19
CD (p = 0.05)		0.28				
200 Gy γ rays +0.2% EMS	7.60 \pm 0.17	+1.35	12.20	9.25	52.69	15.52
300 Gy γ rays +0.2% EMS	7.74 \pm 0.15	+1.49	12.75	9.25	51.02	17.18
200 Gy γ rays +0.3% EMS	6.33 \pm 0.20	+0.08	10.42	6.43	39.53	10.74
300 Gy γ rays +0.3% EMS	5.13 \pm 0.26	-1.12	9.25	4.75	25.91	6.04
CD (p = 0.05)		0.36				

30, respectively. The maximum increase in seeds per pod was recorded in 200 Gy γ rays+0.2% EMS and 300 Gy γ rays+0.2% EMS treatments in the varieties T-9 (3.91) and Pant U-30 (3.94), respectively (Tables 8 and 9). The highest genotypic coefficient of variation (7.10 and 7.45%), and genetic advance (14.21 and 15.74%) was observed with 300 Gy γ rays+0.2% EMS treatment in the varieties T-9 and Pant U-30, respectively in M_2 generation (Table 10). The estimated heritability was maximum with 300 Gy γ rays+0.2% EMS and 200 Gy γ rays+0.2% EMS treatments in the varieties T-9 (63.63%) and Pant U-30 (67.50%), respectively. The results revealed a significant increase in mean seed weight in all the mutagenic treatments in M_3 generation. The maximum increase in mean 100 seed weight was recorded in 300 Gy γ rays+0.2% EMS treatments in the varieties T-9 (4.57) and Pant U-30 (4.63), respectively. The estimated genotypic coefficient of variation was maximum in 200 Gy γ rays+0.2% EMS and 300 Gy γ rays+0.2% EMS treatments in the varieties T-9 (12.03%) and Pant U-30 (12.96%), respectively. The highest heritability (78.95 and 85.71%) and genetic

advance (28.01 and 31.53%) was observed with 300 Gy γ rays+0.2% EMS treatment in the varieties T-9 and Pant U-30, respectively in M_3 generation (Table 10).

100-seed weight is a dependable index for measuring yield ability governed by relatively smaller number of genes unlike other traits. In contrast to the findings of Waghmare and Mehra (2000) in khesari, Singh et al. (2006) in lentil and Giri et al. (2010) in pigeon pea, 100-seed weight, in the present study, had shown a significant increase over the controls with all the mutagenic treatments in M_3 generation. Increase in mean 100-seed weight in the treated population could be attributed to the major incidence of positive mutations.

3.4. Estimates of genetic variability

For all the traits under study, the magnitude of phenotypic coefficient of variation was higher than the magnitude of genotypic coefficient of variation, indicating the role of environment to the total variance

Table 7. Estimates of mean values (\bar{X}), shift in \bar{X} and genetic parameters for number of seeds per pod in M_3 generation of urdbean.

Treatment	Mean \pm S.E.	Shift in \bar{X}	PCV (%)	GCV (%)	h^2 (%)	GA (% of \bar{X})
Var. T-9						
Control	6.22 \pm 0.09	-	7.23	4.18	35.00	6.59
200 Gy γ rays	7.45 \pm 0.12	+1.23	20.24	18.87	85.96	45.50
300 Gy γ rays	7.02 \pm 0.10	+0.80	12.39	11.11	80.26	26.21
CD (p = 0.05)		0.75				
0.2% EMS	7.25 \pm 0.19	+1.03	15.16	13.32	77.31	30.48
0.3% EMS	6.72 \pm 0.21	+0.50	18.20	16.43	81.21	38.84
CD (p = 0.05)		0.44				
200 Gy γ rays +0.2% EMS	7.66 \pm 0.30	+1.44	16.37	14.01	73.08	31.46
300 Gy γ rays +0.2% EMS	7.82 \pm 0.16	+1.60	19.41	17.91	84.85	43.09
CD (p = 0.05)		1.05				
Var. Pant U-30						
Control	6.30 \pm 0.10		7.46	3.81	27.27	5.24
200 Gy γ rays	7.89 \pm 0.17	+1.59	16.08	13.30	68.32	28.89
300 Gy γ rays	7.12 \pm 0.21	+0.82	13.34	11.94	80.00	28.23
CD (p = 0.05)		0.78				
0.2% EMS	7.63 \pm 0.32	+1.33	20.78	19.35	87.95	47.57
0.3% EMS	6.82 \pm 0.19	+0.52	15.25	13.34	76.85	30.64
CD (p = 0.05)		0.47				
200 Gy γ rays +0.2% EMS	7.84 \pm 0.17	+1.54	17.19	15.28	78.02	35.45
300 Gy γ rays +0.2% EMS	8.23 \pm 0.11	+1.93	18.00	16.74	86.76	40.83
CD (p = 0.05)		1.25				

Table 8. Estimates of mean values (\bar{X}), shift in \bar{X} and genetic parameters for 100-seed weight (g) in M_2 generation of urdbean var. T-9.

Treatment	Mean \pm S.E.	Shift in \bar{X}	PCV (%)	GCV (%)	h^2 (%)	GA (% of \bar{X})
Control	3.00 \pm 0.06	-	6.04	3.02	23.07	3.66
100 Gy γ rays	3.28 \pm 0.03	+0.28	7.62	5.25	50.00	10.06
200 Gy γ rays	3.29 \pm 0.02	+0.29	8.20	6.29	57.14	12.46
300 Gy γ rays	3.32 \pm 0.02	+0.32	6.22	4.70	56.82	9.34
400 Gy γ rays	3.10 \pm 0.04	-0.10	7.25	5.28	40.00	7.42
CD (p = 0.05)		0.25				
0.1% EMS	3.14 \pm 0.03	+0.14	8.10	5.48	50.00	10.50
0.2% EMS	3.24 \pm 0.02	+0.24	6.79	5.00	52.08	9.26
0.3% EMS	3.22 \pm 0.02	+0.22	6.52	4.97	59.09	10.25
0.4% EMS	3.01 \pm 0.04	+0.01	7.64	5.40	48.08	9.63
CD (p = 0.05)		0.20				
200 Gy γ rays +0.2% EMS	3.91 \pm 0.03	+0.91	7.20	5.62	61.54	11.51
300 Gy γ rays +0.2% EMS	3.87 \pm 0.04	+0.87	8.54	7.10	63.63	14.21
200 Gy γ rays +0.3% EMS	3.38 \pm 0.01	+0.38	4.44	3.10	45.45	5.32
300 Gy γ rays +0.3% EMS	3.02 \pm 0.05	+0.02	8.30	5.19	41.67	8.94
CD (p = 0.05)		0.62				

(Suganthi and Murugan, 2008). Sharma and Bora (2013) reported similar results for various quantitative characters in vegetable pea. Heritability and genetic advance are important parameters in facilitating the effective prediction of elite genotype selection outcome (Ogunniyan and Olakojo, 2014). Heritability estimates increased for all the traits, except for pod length. Decrease in heritability, in some of the treatments, indicate that even though genetic variance had increased with mutagenic treatment, still the ratio of its increase was not at par with total phenotypic variance exhibited. High heritability was recorded in M_3 as compared to M_2 generation for number of seeds per pod and 100-seed weight, indicating the high probability of their transmission to subsequent generations (Wani and Khan, 2006). Genetic advance differed in different mutagenic treatments in both the varieties. Seeds per pod and 100-seed weight showed comparatively higher genetic advance as compared to pod length, suggesting that these characters are governed by additive gene action (Sharma et al., 2019) and further improvement could be effective through phenotypic selection (Khan and Wani, 2006; Raina et al., 2017). The high heritability estimated for these traits in M_3 generation confirms the induced variance was due to the genotype with least influence from the environment. Considering the large initial mutagenized population, heritability estimates assisted in narrowing down the selection of desirable lines at different treatment conditions and their further

advancement to subsequent M_3 generation. Additionally, the estimates of genetic advance confirmed that the significant genetic gain has been achieved for these traits during the advancement of desirable treatment lines from M_2 to M_3 generation. Owing to polygenic nature of these quantitative traits, the significant statistics obtained from estimation of genetic parameters in M_3 generation confirmed that induced micro-mutations were useful in nature and impacted the yield attributing traits significantly in desirable direction for further selection and advancement.

3.5. Conclusion and future outlook

The mean number of seeds per pod and 100-seed weight had increased with most of the mutagenic treatments in M_2 and M_3 generations. Although pod length did not differ significantly in most of the mutagenic treatments, however few significantly long and bold pods were isolated from individual treatments of EMS in M_3 generation. Long pods can accommodate more seeds with bigger size which could positively impact the net productivity of the crop and might play an important role in creation of new elite genotypes with desirable agronomical traits. Since genetic parameters are useful markers for assessing the genetic diversity of agronomical traits, increased mean values and genetic

Table 9. Estimates of mean values (\bar{X}), shift in \bar{X} and genetic parameters for 100-seed weight (g) in M_2 generation of urdbean var. Pant U-30.

Treatment	Mean \pm S.E.	Shift in \bar{X}	PCV (%)	GCV (%)	h^2 (%)	GA (% of \bar{X})
Control	3.01 \pm 0.02	-	6.38	2.84	18.75	3.01
100 Gy γ rays	3.54 \pm 0.05	+0.53	7.63	4.80	38.89	7.63
200 Gy γ rays	3.73 \pm 0.04	+0.72	8.04	5.79	55.00	11.26
300 Gy γ rays	3.56 \pm 0.02	+0.55	8.42	6.10	57.14	12.36
400 Gy γ rays	3.09 \pm 0.02	+0.08	5.82	4.21	53.12	8.41
CD (p = 0.05)		0.53				
0.1% EMS	3.12 \pm 0.04	+0.11	6.41	4.20	47.22	7.37
0.2% EMS	3.60 \pm 0.02	+0.59	9.72	6.94	50.82	12.78
0.3% EMS	3.39 \pm 0.01	+0.38	7.65	5.90	59.70	11.79
0.4% EMS	3.30 \pm 0.03	+0.29	4.24	2.90	42.10	4.54
CD (p = 0.05)		0.36				
200 Gy γ rays +0.2% EMS	3.68 \pm 0.04	+0.67	5.43	4.60	67.50	9.51
300 Gy γ rays +0.2% EMS	3.94 \pm 0.04	+0.94	9.23	7.45	65.12	15.74
200 Gy γ rays +0.3% EMS	3.32 \pm 0.03	+0.31	5.72	4.20	52.78	7.83
300 Gy γ rays +0.3% EMS	3.15 \pm 0.02	+0.14	6.42	4.20	42.50	6.98
CD (p = 0.05)		0.55				

Table 10. Estimates of mean values (\bar{X}), shift in \bar{X} and genetic parameters for 100-seed weight (g) in M₃ generation of urdbean.

Treatment	Mean \pm S.E.	Shift in \bar{X}	PCV (%)	GCV (%)	h^2 (%)	GA (% of \bar{X})
Var. T-9						
Control	3.44 \pm 0.03	-	5.23	1.60	12.00	1.66
200 Gy γ rays	4.20 \pm 0.06	+0.76	10.24	9.05	77.78	20.71
300 Gy γ rays	4.42 \pm 0.08	+0.98	11.54	10.18	76.92	23.08
CD (p = 0.05)		0.39				
0.2% EMS	4.21 \pm 0.05	+0.77	12.59	10.21	64.28	21.14
0.3% EMS	3.99 \pm 0.04	+0.55	11.28	9.45	70.00	20.80
CD (p = 0.05)		0.53				
200 Gy γ rays +0.2% EMS	4.49 \pm 0.08	+1.05	13.14	12.03	77.14	26.73
300 Gy γ rays +0.2% EMS	4.57 \pm 0.10	+1.13	13.57	11.58	78.95	28.01
CD (p = 0.05)		0.25				
Var. Pant U-30						
Control	3.50 \pm 0.02	-	2.62	1.04	15.00	1.00
200 Gy γ rays	4.25 \pm 0.07	+0.75	11.29	10.12	78.26	23.29
300 Gy γ rays	4.53 \pm 0.09	+1.03	12.14	10.26	70.00	22.52
CD (p = 0.05)		0.28				
0.2% EMS	4.35 \pm 0.06	+0.85	12.18	10.34	71.43	22.76
0.3% EMS	4.11 \pm 0.09	+0.61	13.38	12.16	83.33	29.20
CD (p = 0.05)		0.22				
200 Gy γ rays +0.2% EMS	4.56 \pm 0.10	+1.06	13.38	12.28	83.78	29.38
300 Gy γ rays +0.2% EMS	4.63 \pm 0.12	+1.13	14.04	12.96	85.71	31.53
CD (p = 0.05)		0.67				

diversity of the mutants suggest wider possibilities of selecting more promising urdbean lines with high yield potential.

Declarations

Author contribution statement

Sonu Goyal: Performed the experiments.
 Mohammad Rafiq Wani: Analyzed and interpreted the data; Wrote the paper.
 Aamir Raina; Rafiul Amin Laskar: Analyzed and interpreted the data.
 Samiullah Khan: Conceived and designed the experiments.

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Additional information

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