Radiation induced variability and gene effects for polygenic traits in ricebean (*Vigna umbellata* Thunb, Ohwi and Ohashi)

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The study was conducted to induce variability and to identify the important radiation dose for induction of useful variation for maturity, yield and yield attributes. Two varieties of ricebean (BRS-1 and Totru Local) were treated with three different doses of γ -rays (30kR, 40kR and 50kR). Range, mean and co-efficient of variation suggested that the mutagenic treatments had created wide variability. In general, these genetic parameters were higher in M₃ generation than M₂. Both positive and negative shift in mean were observed for all the traits in both the cultivars in both the generations. High heritability coupled with high genetic advance were observed for pods/cluster, seeds/pod and pod length in both BRS-1 and Totru Local indicating that these traits can respond effectively to phenotypic selection. Most of the traits showed significant positive additive and dominance effects at 30 kR dose in both the genotypes indicating the dose to be most effective in inducing variability. Most of the traits showed overdominance suggesting that the selection should be deferred to the later generation so that the additive effects become more pronounced and fixed.

Key words : γ-rays, Induced additive effect, Induced dominance effect, Mutation, Vigna umbellata

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INTRODUCTION

Ricebean (Vigna umbellata Thunb, Ohwi and Ohashi) is a warm season annual pulse of high nutritional status which has remained in general, neglected for its improvement by breeding interventions either by hybridization or otherwise, especially in India. Till date, it is little known, little researched and little exploited crop. It is being grown sporadically by farmers specifically in the remote areas of Himachal Pradesh and several other states of India. The nutritional quality of ricebean is higher as compared to many other legumes of Vigna family (Katoch, 2013). It has been considered as one of the best nutritionally balanced pulses in the world and has even been included in the school children's nutritional programmes in Philippines (NAS, 1979). Recently, with the emphasis given for the promotion of this crop to be cultivated by Indian farmers, Indian Council of Agricultural Research has started All India Coordinated Research Projects on underutilized crops since 1996 and ricebean is one of them. Under this project, work on the improvement of ricebean has been undertaken by the simplest techniques of germplasm evaluation and hybridization. However, in general, a little work on mutation has been undertaken in this crop. With the increasing awareness and concern among scientists and farmers, ricebean can emerge as an important grain legume crop, provided early, determinate and high yielding varieties are developed.

Variability is the pre-requisite for selection and varietal development in crop plants and mutation induction has become a proven way of creating variation within a crop species. It offers the possibility of inducing desired attributes that either cannot be found in nature or have been lost during evolution. Shu (2009) reported the mutational enhancement of genetic diversity in seventeen plant species. The mutants obtained can become an important genetic resource for breeding, gene discovery and functional analysis of various genes. Among the mutagen used γ -rays are the most widely and effectively used, resulting in about 70 per cent of the worlds' mutant varieties (Nagatomi and Degi, 2009).

As the basic information of components of generated genetic variability is imperative in hybridization, similarly, in case of mutation breeding it is important to know the nature and magnitude of various components of induced genetic variance for planning and executing appropriate breeding strategy to exploit it to the maximum. Also, the nature and magnitude of gene action involved in the expression of traits is important in deciding suitable breeding methodology for population improvement specifically of self fertilizing crops either by hybridization or mutation. Therefore, meaningful genetic analysis of polygenic traits in the mutagenized populations demands for partitioning of hereditary variance into additive, dominance and epistatic components. The present study was, therefore, done to induce variability in two genotypes of ricebean, identify best dose for mutation induction, to know the important characters that respond to mutation and to find out the nature of gene action in ricebean.

Research Methodology

Two contrasting ricebean genotypes *viz.*, BRS 1, having black grains (like urdbean) and high yielding but late maturing, and Totru Local having creamish grains, early maturing but low yielding were used as experimental material. On the basis of LD_{30} and LD_{50} values, three doses of radiations 30kR, 40kR and 50kR were selected for the present study.

Four hundred dry and healthy seeds of each variety were treated with three doses of γ -rays (30kR, 40kR and 50kR). Treatment with γ -rays was done using ⁶⁰Co gamma –cell @2500 R/min installed in the Division of Genetics, at National Physical Laboratory, Indian Agricultural Research Institute, New Delhi. Untreated dry seeds were used as a control.

Sowing of M_1 generation was done immediately after treatment with mutagen during *Kharif* 2006 and single plants were harvested individually and planted as M_2 family row during next crop season. Half of the M_2 seeds of each plant were used for raising M_2 generation *Kharif* 2007 and half were kept for raising M_2 generation *Kharif* 2008 along with M_3 to avoid environmental effect. All M_2 family plots consisted of single five meter long rows with spacing of 45 x 20 cm and 30 x 15 cm for BRS-1 and Totru Local, respectively. Augmented design was used to generate M_3 seeds. Next year, remaining M_2 seeds were sown along with M_3 seeds. M_2 generation was raised in single plant Completely Randomized Design and M_3 generation was raised in Randomized Block Design with three replications.

The number of families studied in BRS-1 were 32, 31 and 29 in M_2 and 33, 27 and 16 in M_3 ; whereas in Totru Local these were 22, 32 and 15 in M_2 and 19, 28 and 11 in M_3 under 30, 40 and 50 kR doses, respectively. Data in M_2 generation were

recorded on single plant basis dose-variety wise, whereas in M_3 generation, the data were recorded on five plants from each line per replication dose-variety wise. Observations were recorded on days to flowering, days to maturity, pod clusters/ plant, pods/cluster, seeds/pod, pod length(cm), seed yield/ plant (g) and 100-seed weight (g).

Estimates of genetic parameters were computed according to Sharma (1998) and the additive and dominance components were estimated according to Yonezawa (1979).

RESEARCH FINDINGS AND ANALYSIS

The findings of the present study as well as relevant discussion have been presented under following heads :

Estimates of genetic parameters :

Analysis of variance for total number of mutagenized populations (1460 in M_2 and 1245 in M_3) exhibited significant differences for all the eight traits studied, indicating the generation of considerable variability after treatment with γ rays. In control no differences were revealed within each parental population for any of the trait, while the two populations as different units differed significantly for all the traits. Homogeneity of individual parental populations indicated that these were most suitable for treatment with mutagens to induce variability for breeding purpose.

Irradiated ricebean varieties exhibited wide range of PCV and GCV for quantitative traits providing ample evidence that mutagenic treatment has altered mean values and created additional genetic variability for quantitative traits. Phenotypic co-efficient of variation were larger compared to genotypic co-efficient of variation for all the traits indicating the influence of environment (Table 1).

During both the generations, high to very high heritability alongwith high to very high genetic advance was observed for pods/cluster, seeds/pod and pod length under 30kR dose in both the varieties. In Totru Local inaddition to above traits pod clusters/plant at 30, pods/cluster at 50kR, seeds/ pod at 40 and 50 kR, pod length at 40 kR and seed yield/plant at 50 kR and in BRS-1, 100-grain weight at 30 kR and pods/cluster at 40 kR were found to be important (these values are in both M_2 and M_3 generation). So, in both the genotypes, 30 kR proved to be the most suitable dose where most of the traits showed moderate to very high PCV, GCV, h_{bs}^2 and GA in both M_2 or M_3 generations (Table 1). Also, the three traits viz., pods/cluster, seeds/pod and pod length have been reported as main yield contributing traits and the data indicated that sufficient variability has been created for further improvement of these traits in both the varieties under 30kR dose. Induced greater variability in polygenic traits might be due to increased mutations and recombinations induced by γ -rays. Increase in the variability parameters in mutagenized populations of ricebean have also been reported by Lokesha

	BRS-1			Totru Local								
Traits	30kR		40kR		50kR		30kR		40kR	50kR		
Generations	M ₂	M ₃	M_2	M ₃								
Days to flowe	ering											
Mean ± SE	76.98 ⁿ	77.54 ^p	77.48 ⁿ	79.29 ^p	76.70 ⁿ	78.69 ^p	69.43 ^p	64.70 ⁿ	69.69 ^p	68.87	70.28 ^p	70.18
	±0.14	± 0.05	±0.18	±0.06	±0.15	±0.10	± 0.18	± 0.10	±0.09	± 0.05	± 0.06	±0.07
PCV	5.93	13.39	13.19	6.28	13.19	8.98	10.88	12.15	4.04	6.40	-	2.71
GCV	5.13	13.23	12.86	5.96	12.86	8.75	10.38	11.90	2.43	5.97	-	1.50
$h_{bs}^{2}(\%)$	74.85	97.67	95.01	89.87	95.05	94.96	91.08	95.93	36.09	87.04	-	30.46
GA	9.14	26.93	25.82	11.63	25.83	17.56	20.41	24.02	3.00	11.47	-	1.70
Days to matu	rity											
Mean ± SE	130.71 ^p	131.00 ^p	131.07 ^p	133.52 ^p	133.23 ^p	135.73 ^p	110.54 ^p	111.51 ^p	109.16 ⁿ	105.18 ⁿ	111.70 ^p	111.89
	± 0.08	±0.30	±0.09	±0.14	± 0.07	±0.20	±0.07	±0.15	±0.06	±0.18	±0.11	±0.25
PCV	6.88	8.78	4.90	6.87	3.36	4.17	4.63	4.03	3.33	5.50	2.90	4.74
GCV	6.75	8.24	4.72	6.20	3.11	2.98	4.36	1.91	2.92	4.02	2.47	3.16
$h_{bs}^{2}(\%)$	96.38	88.18	92.94	81.44	85.72	51.19	88.80	22.53	77.06	53.30	73.02	44.48
GA	13.65	15.95	9.38	11.53	5.94	4.40	8.47	1.87	5.29	6.04	4.36	4.34
Pod clusters/												
Mean ± SE	12.66 ⁿ	11.78 ⁿ	13.95 ^p	13.97 ^p	12.32 ⁿ	12.34 ⁿ	6.89 ⁿ	5.82 ⁿ	7.52 ^p	7.91 ^p	7.52 ^p	7.09 ⁿ
	±0.03	±0.03	±0.03	±0.04	±0.03	±0.04	±0.04	±0.04	±0.04	±0.04	±0.04	±0.04
PCV	11.45	12.30	14.44	11.62	9.76	13.46	46.49	35.08	34.19	17.47	16.35	26.11
GCV	8.59	9.25	12.99	9.39	6.73	11.01	42.86	31.01	32.12	12.62	11.34	22.37
$h_{bs}^2(\%)$	56.35	56.56	80.86	65.37	47.63	66.93	84.97	78.13	88.21	52.20	48.11	73.39
GA	13.29	14.33	24.06	15.64	9.57	18.56	81.38	56.47	62.14	18.79	16.20	39.48
Pods/cluster	13.29	14.55	24.00	15.04	9.57	18.50	01.50	50.47	02.14	10.79	10.20	39.40
Mean \pm SE	4.63 ⁿ	4.49 ⁿ	6.02 ^p	5.74 ^p	5.02 ^p	5.53 ^p	3.29 ⁿ	3.23 ⁿ	4.61 ^p	4.45 ^p	4.01 ^p	3.98 ^p
Mean 1 SE	±0.03	±0.03	±0.04	±0.03	±0.03	±0.04	5.29 ±0.04	5.23 ±0.04	±0.04	±0.03	± 0.05	±0.06
PCV	±0.03 49.60	±0.03 52.41	±0.04 35.05	±0.03 39.89					±0.04 22.12			±0.00 63.10
GCV	49.60 45.47	32.41 47.96	32.30	39.89 36.31	17.26 5.34	33.47 28.73	67.72 61.67	57.12 49.01	22.12 11.90	46.37 41.18	43.11 38.07	
												58.42
$h^2_{bs}(\%)$	84.03	83.74	84.94	82.84	9.58	73.67	82.94	73.61	28.97	78.85	78.00	85.71
GA	85.86	90.41	61.33	68.08	3.41	50.80	115.70	86.62	13.20	75.32	69.27	111.41
Seeds/pod	5 20 B	5.40	< 55 D	< 50 B	5 00 B	5 00 B	4.0.4.1	4.100	4.00 B	4.70 0	4.70 0	2.02
Mean \pm SE	5.38 ^p	5.43	6.55 ^p	6.50 ^p	5.99 ^p	5.89 ^p	4.04 ⁿ	4.13 ⁿ	4.89 ^p	4.70 ^p	4.78 ^p	3.82 ⁿ
DOLL	±0.02	±0.03	±0.03	±0.04	±0.02	±0.04	±0.03	±0.03	±0.05	±0.03	±0.03	±0.05
PCV	29.12	52.07	13.09	24.34	10.68	19.90	39.07	45.83	65.00	46.41	35.11	67.47
GCV	26.37	49.07	7.94	19.53	1.73	11.81	35.00	39.71	63.75	41.84	32.21	62.77
$h_{bs}^{2}(\%)$	81.98	88.82	36.79	64.35	2.63	35.20	80.29	75.09	96.20	81.29	84.15	86.56
GA	49.18	95.27	9.92	32.27	0.58	14.43	64.61	70.89	128.81	77.72	60.86	120.31
Pod length (c												
Mean \pm SE	6.70 ⁿ	6.07 ⁿ	8.01 ^p	7.92 ^p	7.30 ^p	7.03 ⁿ	4.02 ⁿ	4.18 ⁿ	5.31 ^p	5.30 ^p	4.94 ^p	4.41 ⁿ
	±0.05	±0.03	± 0.04	±0.03	± 0.04	±0.06	± 0.04	± 0.04	± 0.05	±0.03	± 0.04	±0.05
PCV	31.36	34.38	18.86	23.55	18.47	19.58	45.83	42.92	31.39	40.91	-	45.64
GCV	27.11	30.82	13.98	20.45	11.39	14.50	36.12	36.77	24.23	36.99	-	40.53
$h_{bs}^{2}(\%)$	74.70	80.35	54.94	75.37	38.04	54.82	62.13	73.40	59.60	81.76	-	78.85
GA	48.26	56.90	21.35	36.57	14.48	22.11	58.66	64.90	38.54	68.90	-	74.13
Seed yield/pla	ιų,											
Mean \pm SE	10.99 ⁿ	11.12 ⁿ	13.01 ^p	13.07 ^p	12.31 ^p	13.10 ^p	6.52 ^p	6.30	7.04 ^p	7.36 ^p	7.24 ^p	6.35
	±0.03	±0.04	±0.04	± 0.05	±0.02	± 0.07	±0.03	± 0.05	± 0.04	± 0.05	± 0.04	± 0.08
PCV	13.67	15.00	13.22	14.44	13.01	10.73	11.35	24.49	15.44	20.44	20.80	31.53
GCV	11.89	10.75	11.96	11.37	11.68	6.03	2.09	16.08	11.62	12.97	18.05	25.67
$h_{bs}^{2}(\%)$	75.68	51.40	81.83	62.04	80.52	31.60	3.40	43.13	56.67	40.24	75.26	66.28
GA	21.30	15.88	22.28	18.45	21.58	6.98	0.79	21.76	18.03	16.94	32.25	43.05
100 seed weig												
Mean ± SE	6.99 ⁿ	6.52 ⁿ	8.32 ^p	7.37 ⁿ	7.73 ^p	7.85 ^p	5.30 ⁿ	4.32 ⁿ	5.86 ^p	5.82 ^p	6.02 ^p	5.14
	±0.05	±0.04	±0.05	±0.04	±0.04	±0.07	±0.04	±0.04	±0.05	±0.04	±0.07	±0.05
PCV	31.01	32.31	23.66	-	14.48	23.51	21.83	60.69	27.48	26.54	-	22.92
GCV	25.93	26.95	19.96	-	5.14	18.26	4.80	54.39	21.92	17.48	-	3.53
$h_{bs}^2(\%)$	23.93 69.92	20.93 69.54	71.15	-	12.60	60.33	4.80	80.33	63.63	43.35	-	2.37
GA	69.92 44.67	69.34 46.29	34.68	-	3.76	29.21	4.85 2.17	80.33 100.42	36.02	43.33 23.71	-	1.12

p = Significant positive shift in mean

n = Significant negative shift in mean



et al. (1991) and Lokesha and Veeresh (1993).

Estimates of mean values of irradiated populations :

In BRS-1, at 30 kR dose at M₂ generation, five traits showed significantly negative shift in mean values while, in Totru Local six traits shifted negatively. In BRS-1, days to flowering showed significantly negative shift in mean in M₂ generation and significantly positive shift in M₃ generation under all the doses (Table 1). The increase values could be due to the occurrence of polygenic mutations with cumulative effects. In Totru Local the shift towards earliness was shown only at 30kR dose in M₂ generation.

Since for days to flowering and maturity negative value (earliness) is of interest for the breeders so, a shift in positive direction is required for fulfillment of Brock's hypothesis

(1965). For days to flowering, Brock's hypothesis fitted well in M₂ generation under all the doses in BRS-1 and in Totru Local all doses except at 30 kR. For days to maturity, hypothesis fitted well for all the doses in both the varieties except for 40kR in Totru. So, the possibility of getting early mutants in Totru local under 40kR dose can be speculated.

The above results showed 30 kR to be the dose where most of the traits shifted their mean values from the previous generation. Similar shift in mean were also reported by Kharkwal (2001) in chickpea and Singh et al. (2006) in cowpea.

Genetic component analysis of induced polygenic variation :

Significant and positive additive and dominance effects were observed for all the traits except for days to maturity and seeds/pod at 30kR treatment; for 100 seed weight at 40kR and

Dose / Traits	Days to flowering	Days to maturity	Pod clusters/plant	Pods/cluster	Seeds/pod	Pod length (cm)	Seed yield /plant (g)	100 seed weight (g)
BRS-1								
30 kR								
(d) _m	$-0.43* \pm 0.09$	$\textbf{-0.56}{\pm0.30}$	$1.14^{*\pm} 0.04$	$0.28^{\pm} \pm 0.04$	-0.04 ± 0.04	$0.82*\pm0.04$	$0.21*\pm0.05$	$0.71^{*\pm} 0.06$
[ĥ] _m	$-1.12^{\pm} \pm 0.30$	$\textbf{-0.58}{\pm0.62}$	$1.76^{*}{\pm}~0.08$	$0.28^{*}{\pm}~0.08$	-0.10 ± 0.07	1.26*±0.12	0.26*±0.10	0.94*± 0.13
$[\hat{\mathbf{h}}]_{\mathbf{m}}$ / $[\hat{\mathbf{d}}]_{\mathbf{m}}$	2.60	-	1.54	1.00	_	1.53	1.24	1.33
40 kR								
[â] _m	$-1.93* \pm 0.11$	$-2.91*\pm 0.15$	$-0.41* \pm 0.05$	$-0.28* \pm 0.04$	$-0.52*\pm0.05$	-0.37*±0.04	-0.73*±0.06	$0.52* \pm 0.06$
[ĥ] _m	$-3.62* \pm 0.38$	$-4.90* \pm 0.33$	-0.04 ± 0.10	$0.56^{*}{\pm}~0.10$	0.10±0.10	0.18±0.10	-0.12±0.13	$1.90* \pm 0.13$
[ĥ] _m /[d̂] _m	1.88	1.69	-	2.04	_	-	-	3.65
50 kR								
[â] _m	$-1.72^{*\pm} 0.13$	$-4.04* \pm 0.21$	$0.41* \pm 0.05$	-0.57*±0.05	-0.19*±0.05	0.17*±0.07	$-1.11*\pm0.07$	$-0.27*\pm 0.08$
[ĥ] _m	$-3.98* \pm 0.36$	$-5.00* \pm 0.42$	-0.04 ± 0.10	$-1.02*\pm0.10$	0.20*±0.09	0.54*±0.14	-1.58*±0.15	-0.28 ± 0.16
[ĥ] _m /[d̂] _m	2.31	1.24	-	1.81	1.05	3.27	1.42	-
Fotru Local								
30 kR								
[â] _m	$4.49^{\pm} \pm 0.14$	$-1.18* \pm 0.16$	$1.24^{*\pm} 0.05$	$0.38^{*}{\pm}~0.05$	-0.02 ± 0.04	0.16*±0.05	0.13*±0.05	$1.10^{*} \pm 0.05$
[ĥ] _m	$9.46^{\pm} \pm 0.41$	$-1.94* \pm 0.33$	$2.14* \pm 0.11$	0.12 ± 0.11	-0.18*±0.08	-0.32*±0.11	0.44*±0.12	$1.96^{*\pm} 0.11$
[ĥ] _m /[ẩ] _m	2.11	1.65	1.73	_	_	2.06	3.52	1.79
40 kR								
[â] _m	$0.45^{\pm} \pm 0.07$	$4.46^{\pm} \pm 0.19$	$-0.54* \pm 0.05$	-0.19*±0.04	$0.17*\pm0.05$	-0.32*±0.05	-0.68*±0.05	-0.13*± 0.05
[ĥ] _m	$1.64* \pm 0.21$	$7.96^{*\pm} 0.38$	$-0.78^{*\pm} 0.11$	0.32*±0.10	0.38*±0.12	0.02 ± 0.12	-0.64*±0.13	0.18 ± 0.13
$[\hat{\mathbf{h}}]_{\mathbf{m}}$ / $[\hat{\mathbf{d}}]_{\mathbf{m}}$	3.64	1.78	1.46	1.73	2.30	_	0.95	_
50 kR								
[â] _m	$-0.56^{*} \pm 0.08$	$-0.98* \pm 0.26$	$0.29^{*\pm} 0.05$	-0.02 ± 0.07	0.66*±0.06	$0.39* \pm 0.06$	0.44*±0.08	$0.64* \pm 0.07$
[ĥ] _m	0.20 ± 0.18	-0.38 ± 0.55	$0.86^{*\pm} 0.11$	0.06±0.16	1.92*±0.12	$1.06^{*\pm} 0.13$	1.78*±0.18	$1.76^{*} \pm 0.17$
[ĥ] _m /[d̂] _m	_	_	3.02	_	2.91	2.75	4.09	2.77

* indicate significance of values at P=0.05 respectively

Asian J. Bio Sci., 9 (1) April, 2014 : 53-57 Hind Institute of Science and Technology



for pod length at 50kR in BRS-1 (Table 2). In Totru Local, positive and significant additive and dominance effects along with potent ratio in the range of over dominance were observed for days to flowering, pod clusters/plant, seed yield/plant and 100 seed weight in 30 kR; days to flowering, days to maturity and seeds/pod in 40kR and pods cluster/plant, seeds/pod, pod length, seed yield/plant and 100 seed weight in 50 kR. Virk et al. (1978) have attributed the induction of such positive additive effects due to fact that either decreasing alleles have high mutation rate or the parents contain more increasing alleles than decreasing one for the traits exhibiting positive additive effect. Jai Dev and Gupta (1997) in rajmash and Pathania et al. (2011) in chickpea also reported predominance of both additive and dominance genetic effects for most of the traits after irradiation with γ -rays.

Also, under 30kR dose, significant additive and dominance effects were observed for two yield component traits (pod clusters/plant and 100-seed weight) and other yield contributing trait (days to flowering) in both the genotypes. Considering all the doses in both the genotypes, it was observed that most of the traits showed overdominance which suggested that the selection should be deferred to the later generation so that the additive effects become more pronounced and fixed.

In the present study the 30kR dose have come out to be effective in inducing considerable variation for a number of characters of economic value. Yield contributing characters like pods/cluster, seeds/pod and pod length showed high hertitability and genetic advance at one or other dose of mutagens in both the varieties. Therefore, the variation induced in these characters could pave the way for selection of high yielding mutants for further studies in late generation when the additive effects will become more pronounced and fixed.

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