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Morphological changes and induced mutagenesis in gladiolus varieties through gamma irradiation

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ABSTRACT

An experiment was conducted for 3 years (2011-12 to 2013-14) at Varanasi to find out influence of various gamma doses, i.e. 1.5, 2.5, 3.5, 4.5 and 5.5 kR along with untreated (control) in different varieties of gladiolus, viz. Aldebaran, Jyotsana, Legend, Praha, Punjab Dawn, Pusa Kiran and Tiger Flame. During 1st and 2nd years various parameters were observed and data were pooled for statistical analysis, whereas during 3rd year stability of characters was observed among mutants. The experiment was laid out in randomized block design with 5 replications. Maximum leaf area was observed with control plants followed by 1.5 kR, 2.5 kR, 3.5 kR, 4.5 kR and 5.5 kR treatments. Narrower and short leaves were observed as gamma doses increased. Maximum length of spike at 1st floret opening was recorded with control followed by 1.5, 2.5, 3.5, 4.5 and 5.5 kR. There was no significant difference between 1.5 kR and 2.5 kR treatments on length of spike at 1st floret opening. Gamma dose at 2.5 kR registered durability of 1st floret followed by 1.5 kR, control, 3.5 kR, 4.5 kR and 5.5 kR treatments. No significant difference was observed between 2.5 kR and 1.5 kR treatments. Control plants gave maximum diameter of 3rd floret followed by 1.5 kR, 2.5 kR, 3.5 kR, 4.5 kR and 5.5 kR. Among varieties, cv. Jyotsana registered maximum length of spike, cv. Punjab Dawn resulted in maximum durability of floret and cv. Tiger Flame registered maximum diameter of 3rd floret. Interaction of gamma doses and varieties were also found significant in all the characters studied. Various morphological changes were observed on gladiolus varieties in cv. Jyotsana, Legend and Praha at 1.5 to 3.5 kR doses of gamma. A mutant was identified from variety Tiger Flame at 4.5 kR of gamma irradiation. This mutant was observed during 1st and 2nd years. It was a stable mutant, hence, isolated during 3rd year. Flower colour of mutant was different to the parent. Lower petals of mutant were found of yellow colour. It looks very attractive.

Key words: Flowering, Gladiolus, Gamma irradiation, Mutant

Among the commercial flowers, gladiolus is one of the most important flowers in India because of its majestic spikes containing attractive, elegant and delicate florets of various shades, sequential opening of flowers for a longer duration and good keeping quality of cut spikes (Singh 2006). The growing of gladiolus especially the multicoloured varieties are increasing day by day. Varying in different colours, sizes and long keeping quality of flower spikes make this crop very popular in domestic market. Thus, possesses a great potential for export market especially during winter. These have mostly been evolved through conventional breeding but a few through mutation breeding. Ornamental plants are ideal for the application of mutation induction techniques because many characters of economic interest, i.e. flower traits (novelty, doubleness, petaloides, dwarfness, vase life, leaf variegation, biotic and abiotic resistance) are easily monitored after the mutagenic treatment. The literature cited by Broertjes and van Harten (1988) and the publications during the past 10 years show that 55% of the records concerned changes in flower colour and 15% in flower morphology. Raghava *et al*. (1988) studies the effect of gamma rays from ^{60}Co was in three varieties of gladiolus namely Little Giant, Mansoer and Wild Rose that were irradiated at 1, 2.5, 5, 10 and 15 kR doses. Observations reported that percentage of sprouting was affected significantly at 10 and 15 kR. LD_{50} was found to be between 10 and 15 kR. Gladiolus mutant are generally lighter in colour than the original parent. Cultivar white friendship is a very popular mutant obtained from its known parent friendship. IIHR, IARI, NBRI and other organization did some significant contribution in this line. Various workers have been adopted different doses of gamma irradiation for improvement of gladiolus. In general higher doses were found to be lethal and doses from 0.5 to 5.0 krad are

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advisable for the gamma irradiation in gladiolus (Singh 2014). Hence, present investigation was carried out by applying lower doses of physical mutagens in different varieties of gladiolus.

MATERIALS AND METHODS

The present investigation was carried out for 3 years during 2011-2012, 2012-2013 and 2013-2014 at Horticulture Research Farm and Post-harvest Laboratory, Department of Horticulture, Banaras Hindu University, Varanasi, Uttar Pradesh. Gamma doses at 1.5 kR, 2.5 kR, 3.5 kR, 4.5 kR, 5.5 kR including control were exposed to 7 varieties, viz. Aldebaran, Jyotsana, Legend, Praha, Punjab Dawn, Pusa Kiran and Tiger Flame of gladiolus corms at National Botanical Research Institute, Lucknow. These treated corms were planted in the beds with a spacing of row to row 30 cm and corm to corm 20 cm during November 2011. 2nd and 3rd generations corm were also planted in same manner. The experimental site lies approximately in the centre of North-Gangetic alluvial plain; on the left bank of river Ganga is homogeneously fertile with uniform textural makeup. Varanasi city situated at 25º 10' north latitude and 83º 03' East longitudes. The altitude is 123.23 meter above the mean sea level. The climate of Varanasi is humid subtropical with large variation between summer and winter temperature. During the experiment the maximum temperature ranged from 14.2ºC to 43.0ºC in summer and minimum temperature 7.1ºC to 28.1ºC in winter. During 1st and 2nd years various parameters were observed and data were pooled for statistical analysis, whereas during 3rd year stability of characters was observed among mutants. The experiment was laid out in randomized block design with 5 replications. Various parameters were observed on growth and flowering, i.e. leaf area, length of spike at first floret opening, durability of 1st floret and diameter of 3rd floret. Morphological changes were observed in some varieties due to gamma doses. A stable mutant was identified and isolated from parent. Details of this colour mutant are described with the colour chart developed by Royal Horticulture Society, London.

RESULTS AND DISCUSSION

Leaf area

Data pertaining to the leaf area found statistically significant due to gamma doses, varieties and their interactions (Table 1). Maximum leaf area was observed with control plants followed by 1.5 kR, 2.5 kR, 3.5 kR, 4.5 kR and 5.5 kR treatments it was significantly higher than all the doses of gamma irradiation. No significant difference was observed in leaf area of 1.5 kR and 2.5 kR treatments and minimum leaf area was recorded with higher doses of gamma irradiation (5.5 kR treatment). It is imperative to state that as doses of gamma irradiation increased leaf area of plants decreased. It is indicating that normal leaves produced in control plants, whereas leaf's morphology changed in case of all treated plants and narrower and short leaves were observed as gamma doses increased, hence, leaf area decreased in general due to various doses of gamma irradiation. Among cultivars, var. Tiger Flame registered maximum leaf area followed by cv. Jyotsana, Punjab Dawn, Pusa Kiran, Aldebaran and Legend. Interaction of 1.5 kR treatment and cv. Jyotsana registered maximum leaf area, whereas it was minimum with interaction of 5.5 kR treatment and cv. Legend. Findings of earlier workers are also incongruence with present work. Morphological changes including abnormalities in the foliage also observed in the irradiated material of gladiolus (Singh and Kumar 2013). Various qualitative changes observed on the gladiolus plants and flowers in variety Pusa Kiran (Sisodia and Singh 2014).

Table 1 Effect of gamma irradiation on leaf area (cm^2) and length of spike at the opening of 1st floret (cm) in the different varieties of gladiolus

Variety	Leaf area (cm^2)							Length of spike at the opening of 1st floret (cm)						
	Treatment							Treatment						
	Control	1.5 kr	2.5 kr	3.5 kr	4.5 kr	5.5 kr	Mean	Control	1.5 kr	2.5 kr	3.5 kr	4.5 kr	5.5 kr	Mean
Aldebaran	71.92	80.17	78.39	66.78	54.93	50.55	67.12	62.77	71.23	68.16	30.64	27.20	31.44	48.58
Jyotsana	86.41	110.87	105.04	102.00	88.93	41.65	89.15	80.32	60.14	68.92	69.54	88.24	25.6	66.79
Legend	94.01	40.31	44.83	38.55	30.24	32.75	47.05	62.13	30.44	31.50	0.00	0.00	0.00	21.79
Praha	79.75	80.03	81.66	66.55	55.86	70.17	72.34	53.63	59.82	54.60	30.63	20.57	26.71	40.01
Punjab Dawn	106.61	97.39	81.21	80.31	84.40	33.63	80.59	69.67	67.47	64.05	52.52	49.75	24.94	54.73
Pusa Kiran	94.38	84.80	82.10	78.92	70.40	68.92	80.24	66.94	61.34	71.69	29.88	24.60	23.10	46.26
Tiger Flame	93.23	102.65	105.16	106.68	100.20	47.48	93.06	82.37	71.68	72.41	64.90	67.05	11.24	61.61
Mean	89.47	85.17	82.75	77.11	69.28	49.30		68.26	61.80	61.13	39.73	39.84	20.43	
CD (P=0.05)														
Treatment				3.70							3.07			
Variety				3.99							3.31			
Treatment × Variety				9.80							8.13			

Length of spike at 1st floret opening

Different doses of gamma irradiation and varieties showed significant effect on length of spike at first floret opening (Table 1). Maximum length of spike at 1st floret opening was recorded with control (68.26 cm) followed by 1.5 kR, 2.5 kR, 3.5 kR, 4.5 kR and 5.5 kR. Whereas, no significant difference was observed between 1.5 kR and 2.5 kR doses of gamma irradiation. There was no significant difference between 4.5 kR and 5.5 kR treatments on length of spike at 1st floret opening. It was noticed that at doses higher doses of gamma irradiation (5.5 kR) length of spike decreased significantly. Length of spike at first floret opening was influenced by different varieties. Cultivar Jyotsana registered maximum length of spike (66.74 cm) at 1st floret opening followed by cv. Tiger Flame, Punjab Dawn, Aldebaran, Pusa Kiran and Praha which was statistically superior to other varieties. Interaction of 4.5 kR treatment with cv. Jyotsana resulted in maximum length of spike. The reason for the reduction in spike length and floret size might be due to inhibition of growth. Most of the plants remain in the juvenile stage and whether unable to produce spike or differentiate floral bud. At higher doses effect of gamma irradiation was more pronounced which resulted in smaller spike length and reduced flower size. This might be due to reduction in plant growth. Banerji and Datta (2002) also reported similar findings while working with chrysanthemum cv. Lalima after gamma irradiation. Reduction in stem diameter, plant spread and plant height was also noticed by Singh *et al*. (2009) in marigold due to irradiation of seeds in African marigold cv. Pusa Narangi Gainda.

Durability of 1st floret in the field

It is apparent from the Table 2 that durability of 1st floret influenced significantly due to gamma irradiation, varieties and their interaction. Maximum durability of 1st floret (5.71 days) was observed with 2.5 kR treatment followed by 1.5 kR, control, 3.5 kR, 4.5 kR and 5.5 kR treatments. No significant difference was observed between 2.5 kR and 1.5 kR treatments, whereas durability of 1st florets in these treatments were statistically higher than control. However, durability of 1st florets was significantly lower in 3.5 kR, 4.5 kR and 5.5 kR treatments. Among the varieties maximum durability was recorded with cv. Punjab Dawn (6.32 days) followed by cvs. Jyotsana, Pusa Kiran, Tiger Flame, Praha, Aldebaran and Legend. Durability of 1st florets of cv. Punjab Dawn was significantly higher than other varieties. Interaction of 1.5 kR treatment with cv. Jyotsana gave spectacular response on durability of 1st floret which was statistically higher than other treatment combinations. These results are experimentally substantiated with the observation made by Patil *et al*. (2010) and Patil (2011) who noticed that lower dose of gamma irradiation found beneficial in improving some floral parameters and higher dose found injurious for most of the flowering parameters in gladiolus.

Diameter of 3rd floret

Various doses of gamma irradiation and varieties influenced significantly on diameter of 3rd floret (Table 2). Among various treatments, control plants gave maximum diameter of 3rd floret followed by 1.5 kR, 2.5 kR, 3.5 kR, 4.5 kR and 5.5 kR. No significant difference was observed between 1.5 kR and 2.5 kR. In case of higher gamma doses (3.5 kR, 4.5 kR and 5.5 kR), size of 3rd floret reduced significantly. Among varieties, cv. Tiger Flame registered maximum diameter of 3rd floret (8.51 cm) followed by cv. Jyotsana, Punjab Dawn, Praha, Pusa Kiran, Aldebaran and Legend. Interaction of 2.5 kR with cv. Tiger Flame registered maximum diameter of 3rd floret. The present findings in many respects are in tune with the observation of Misra and Bajpai (1983) who noted that gamma rays proved favourable at lower doses and when corms of three varieties subjected

Table 2 Effect of gamma irradiation on durability of 1st floret (days) and diameter of 3rd floret (cm) in the different varieties of gladiolus

Variety	Durability of 1st floret (days)							Diameter of 3rd floret (cm)						
	Treatment							Treatment						
	Control	1.5 kr	2.5 kr	3.5 kr	4.5 kr	5.5 kr	Mean	Control	1.5 kr	2.5 kr	3.5 kr	4.5 kr	5.5 kr	Mean
Aldebaran	3.30	3.60	4.00	2.30	2.50	2.40	3.02	9.75	5.29	4.67	4.21	4.40	3.96	5.37
Jyotsana	3.50	7.00	6.60	6.00	5.10	2.00	5.03	7.73	9.43	8.96	9.65	9.84	3.25	8.14
Legend	6.00	2.80	5.50	0.00	0.00	1.93	2.70	7.69	4.30	3.85	0.00	0.00	0.00	2.64
Praha	5.00	5.00	4.20	2.10	1.80	2.00	3.35	8.99	9.25	9.39	4.22	3.91	4.30	6.68
Punjab Dawn	5.20	6.90	7.85	6.10	5.40	6.60	6.32	8.99	8.59	9.28	7.92	8.15	3.10	7.67
Pusa Kiran	5.40	6.20	5.90	2.50	2.7	5.00	4.61	8.29	8.97	9.09	4.53	4.15	3.90	6.46
Tiger Flame	5.30	6.20	6.10	4.60	4.90	2.00	4.85	9.86	9.51	10.08	8.85	8.47	4.30	8.51
Mean	4.81	5.30	5.71	3.37	3.22	3.13		8.75	7.90	7.90	5.62	5.56	3.25	
CD (P=0.05)														
Treatment				0.44							0.34			
Variety				0.47							0.37			
Treatment × Variety				1.18							0.92			

to treat with gamma doses and LD_{50} proved at higher doses which affect growth and flowering traits. It was also noticed that sensitivity of radiation varied between varieties and between traits. Present findings also corroborated with the observation of Rather and Jhon (2000) who noticed that spike length and flower size reduced when bulb of iris were treated with gamma doses.

Morphological changes in gladiolus varieties Jyotsana, Legend and Praha

Various morphological changes were observed on gladiolus varieties. In cv. Jyotsana abnormal growth of spike was observed at 1.5 kR dose (Fig 1), whereas at 3.5 kR gamma irradiation leaves were wider and stunted spikes were emerged in cv. Jyotsana (Fig 2). In cv. Legend abnormal spike with single floret was observed at 2.5 kR gamma dose (Fig 3). Whereas, at same dose a plant emerged spike in which florets were arranged in both direction in var. Legend (Fig 4). In cv. Praha faded red colour was observed at 1.5 kR dose. On same plant purple blotches were observed on flowers (Fig 5). Similar to cv. Legend, at 2.5 kR dose of gamma irradiation florets on both sides were appeared in cv. Praha (Fig 6). These chimeras and mutants were not observed in 3rd year (VM_3) of observation. Morphological changes in gladiolus were also noticed by earlier workers. Jakota and Murin (1994) obtained mutants with white spots on tepals during study for the genetic variability of gladioli as the result of gamma radiation. However, on studying the effect of gamma irradiation on cvs. Green Finch, Mayur, Rose Momento and Wind Song, colour variations were recorded even from lowest dose of 1.5 kR (Misra 1996). They also reported changes in colour of petals, fusion and fasciation,

Fig 1 1.5 kR, abnormal growth of spike, cv. Jyotsana

Fig 2 3.5 kR, more width of leaf, stunted spike, cv. Jyotsana

Fig 3 2.5 kR, broad leaves, abnormal spike, single floret, cv. Legend

Fig 4 2.5 kR, florets in opposite direction at same node, cv. Legend

Fig 5 1.5 kR, fading of red colour, purple blotches cv. Praha

Fig 6 2.5 kR, florets on both sides cv. Praha

Table 3 Characteristic of gladiolus matant (4.5 kR gamma dose) along with parent Tiger Flame

Characters	Parentage	Mutant
Length of leaf	38.7 cm	54.2 cm
Width of leaf	3.7 cm	3.4 cm
Width of scape	6.2 cm	4.4 cm
Number of leaves	7	7
Rachis length	58.6 cm	59.5 cm
Number of florets/ spike	16	16
Length of floret	10 cm	10.5 cm
Width of floret	8.25 cm	9.0 cm
Floret colour	Deep orange	Upper petal light orange with streaks
Throat colour	Orange with shade of yellow	Lower petal yellow, no throat
Length of 1st petal	6.3 cm	7.5 cm
Length of 2nd petal	6.8 cm	7.0 cm
Length of 3rd petal	5.5 cm	6.0 cm
Length of 4th petal	8.3 cm	8.5 cm
Length of 5th petal	9.0 cm	9.3 cm
Length of 6th petal	8.0 cm	8.2 cm
Width of 1st petal	3.5 cm	3.5 cm
Width of 2nd petal	2.7 cm	2.1 cm
Width of 3rd petal	2.5 cm	2.2 cm
Width of 4th petal	4.5 cm	4.5 cm
Width of 5th petal	4.5 cm	4.7 cm
Width of 6th petal	4.5 cm	4.7 cm
Pistil length	7.3 cm	7.9 cm
Stamen length	4.7 cm	4.7 cm
Petal 1	Upper portion-Orange red 30 A Lower portion-green yellow 1CThroat-orange 25 C	Upper portion-orange red 32 ALower portion-yellow 6 DThroat (streak like)-red purple N 57 B
Petal 2	Upper portion-orange red C Lower portion-yellow 2 BThroat-orange N 25 A	Upper portion-yellow 6 DLower portion-orange red 32 A No throat
Petal 3	Upper portion-orange red C Lower portion-yellow 2 BThroat orange N 25 B	Upper portion-yellow 6 DLower portion-orange red 32 A No throat
Petal 4	Orange red 33 B	Orange red 32 B
Petal 5	Orange red 33 B	Orange red C
Petal 6	Red 40 B	Orange red 32 B

increased floral organ, in all the varieties. Kasumi *et al*. (1999) found a mutated sectorial chimeric flower colour variant obtained by gamma irradiation of cormels of gladiolus cv. Traveler. Morphological changes in the flowers were also observed due to gamma irradiation in gladiolus (Singh and Kumar 2013).

Induced mutant

A stable mutant was isolated from variety Tiger Flame at 4.5 kR of gamma irradiation. This mutant was observed during 2011-2012 experimentation (1st year) and persisted to 3rd generation (VM_3) during 2013-2014. Flower colour of mutant was different from the parent. Distinct difference was colour of lower petals of flower. In mutant lower petals were of yellow colour (Table 3). In general, size of flower was larger than parent (Table 3). Distinct changes were noticed when individual petal study was carried out with the help of flower colour chart developed by The Royal Horticultural Society, London. The lower portion of 1st petal mutant was of yellow colour, whereas, upper portion of petal of parent was orange red (32 A) in colour. Throat was absent in mutant, however, it was present in the 1st petal of the parent (orange 25 C). In parent upper portion of the 1st petal was of orange red (30 A) colour. Green yellow (8 C) was found on the lower portion of the petal. On 2nd petal of mutant yellow colour (6 D) was observed on lower portion of petal and upper portion was of orange red (32 A). Throat was absent, whereas, on 2nd petal of parent throat was present of orange (N 25 A) colour. Lower portion of 2nd petal of mutant was yellow (2 B) colour. Upper portion was of orange red (C) colour. Similarly, such difference between mutant and parent was also observed on 3rd petal. 4th, 5th and 6th petals of mutant were orange red colour with various shades i.e, 32 B, C and 32 B respectively. Whereas, in parent 4th and 5th petals was of orange red (33 B) colour and red colour (40 B) was observed in 6th petal. Difference on petals (1-6 petals) size was also observed between parent and mutant as given in Table 3. Misra (1982) detected colour mutant in gladiolus cultivar Ratna's Butterfly. In the mutant all the segments in upper half have violet colour with lighter flakes in between. A desirable and stable mutant isolated from the variety Wild Rose in 1 kR treatment and was released as Shobha (Raghava *et al*., 1988) while studying the effect of gamma rays from ^{60}Co. in three varieties of gladiolus namely Little Giant, Mansoer and Wild Rose irradiated at 1, 2.5, 5, 10 and 15 kR doses.

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