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Biological Effects of Gamma Irradiation and its Modulation with Gibberellic Acid in M_1 Generation of Chickpea (*Cicer arietinum* L.)

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Abstract: Dry seeds of three chickpea genotypes having different seed coat colours viz; Noor 91 (white), Punjab 91 (brown) and C 141 (black) were treated at 40, 50 and 60 Kr separately and with gibberellic acid (GA_3) for studying the effects on 100-seed weight, grain yield, biological yield, harvest index, days to flowering and maturity in M_1 generation. Highly significant ($p < 0.01$) variation within genotypes, treatments and also for their interaction was observed for all the characters. Statistically higher 100-seed weight was observed with the combine treatment in the three genotypes as compared with gamma irradiation. Combine treatment normalized the grain yield in Noor 91, while decreased in Punjab 91 however, stimulation was recorded at 40 and 50 Kr in C 141. Stimulation in biological yield was recorded at 40 Kr with the combine treatment in Noor 91 and C141. Harvest index decreased with gamma irradiation in the three genotypes while, stimulation was recorded in C 141. Days to flowering and maturity increased with gamma irradiation however, with combine treatment these values were reduced.

Key words: Gamma irradiation, modulation, gibberellic acid, Chickpea, *Cicer arietinum*, radiosensitivity

Introduction

Mutation breeding is now widely used for inducing genetic changes and creation of new genetic resources, particularly in crops that are not easily amenable to improvement through conventional techniques (Awan, 1991). It is well established that mutagenic agents are effective for inducing genetical changes in treated population (Kalia and Gupta, 1988a, b). Notwithstanding, little natural variability in chickpea for conspicuous morphological and physiological characters, several workers have attempted for induction of mutation using either physical or chemical mutagens for evolving new genotypes (Hassan and Khan, 1991; Shamsuzzaman and Shaik, 1991). Radiation, therefore, appears to be a useful tool in plant breeding and genetics. The primary objectives of mutation are to enhance mutation frequency, widen the mutation spectrum and realize directed mutagenesis.

Gamma radiation in combination with other chemical mutagens is applied for widening the frequency and mutation spectrum for extra genetic variability. Effect of gamma radiation is changed with the radio protective effect of gibberellic acid. It has been established that the impaired growth due to gamma irradiation can be restored by exogenous application of gibberellic acid. Gibberellic acid serves manifold growth related functions in plants by enhancing replication, transcription and different enzymatic systems (Zhebrak, 1989; Ali and Ansari, 1989; Arora *et al.*, 1989). It brightens the scope for increasing both the frequency and spectrum of mutation. Therefore, it was planned to determine the effectiveness of gamma irradiation and efficiency of gibberellic acid to modulate the radio sensitivity for various morphological characters.

Materials and Methods

Dry seeds were exposed to gamma irradiation at doses of 10, 20, 30, 40, 50, 60, 70, 90 and 110 Kr to 1000 seeds for each treatment in three genotypes at Nuclear Institute for Food and Agriculture (NIFA), Peshawar. A part of the irradiated seeds after one hour of soaking under continuous aeration, were subjected to 0.5 mM aqueous solution of gibberellic acid for 16 hours with constant shaking. Non irradiated seeds soaked in water were kept as control in the case. After treatment seeds were washed in running tap water and then were dried on blotting paper. On the basis of seedling performance doses of 40, 50 and 60 Kr were selected for inducing genetic variability on large scale. Treated along with control seeds were sown in split plot design with three replications at Barani Agriculture Research Institute (BARI) Chakwal in 1995 to raise the M_1 generation. Data on 20 consecutive plants from the middle row was appropriately recorded for various characters.

Results and Discussion

The analysis of variance for the effect of different doses of gamma irradiation separately and with the post mutagenic application of gibberellic acid on various plant characteristics in M_1 population of chickpea is presented in Table 1. It indicates highly significant variation within genotypes and treatments for all characters. The genotype-treatment interaction was also highly significant for all the characters. It reflects highly inconsistency in sensitivity of genotypes for all the characters across various treatments.

100-seed weight decreased significantly with an increase in gamma irradiation (Table 2). In the previous research, similar observations for this character was recorded in chickpea (Rao, 1988), in mungbean (Khan, 1984) and in *Pennisetum* (Aslam *et al.*, 1985). However, heavier seeds were produced with the application of GA_3. This increase in 100-seed weight may be due to the radio protective effects of GA_3 and enhancement of template activity. The grain yield reduced significantly at various levels of irradiation. Similar results for this character was obtained in various crops; in chickpea (Mahto *et al.*, 1989), in lentil (Eser *et al.*, 1991; Tripathi and Dubey, 1992), in *Pennisetum* (Aslam *et al.*, 1985), in *Cajanus cajan* (Kumar and Sinha, 1989) and in *Phaseolus vulgaris* (Svetleva and Dimeva, 1991). However, with the application of GA_3 grain yield increased significantly at 40 and 50 Kr and decreased at 60 Kr. Grain yield decreased significantly or non-significantly with gamma irradiation in the three genotypes as compared to their respective controls. However, the response of genotypes varied greatly at different doses. Genotypic differences due to various gamma irradiation were also observed by Mahto *et al.* (1989) in chickpea and Aslam *et al.* (1985) in *Pennisetum.* Application of GA_3 had changed the effects of gamma irradiation significantly in the three genotypes except at 60 Kr in Punjab 91 indicating the possibility of increasing the variability for grain yield in chickpea.

Both the mutagenic treatments decreased the biological yield

Table 1: Mean of Squares for different characters of M_1 generation in chickpea genotypes.

Variables	D.F.	100-Seed Weight	Grain yield/plant	Biological Yield/plant	Harvest Index	Days to Flowering	Days to Maturity
Replicates	2	0.054	0.92	9.05	0.54	0.28	1.29
Varieties (V)	2	4.90**	487.52**	920.13**	315.84**	119.28**	101.16**
Error a	4	0.037	1.58	2.04	7.52	0.04	0.21
Treatments (T)	7	4.345**	143.63**	401.28**	82.08**	175.10**	93.29**
TxV	14	0.121**	109.47**	182.80**	89.20**	4.38**	4.43**
Error b	42	0.023	1.342	2.63	0.44	0.36	0.30
Total	71						

** 0.01 probability highly significant

Table 2: Effect of gamma irradiation separately and with gibberellic acid on various characters in M_1 generation of three chickpea genotypes.

		Control		Gamma irradiation			Gamma irradiation +GA_3 (0.5 mM)			
Variables	Varieties	Dry	Soaked	40 Kr	50 Kr	60 Kr	40+ GA_3	50+ GA_3	60+ GA_3	Mean
	Noor 91	25.10 a	25.02 a	24.77 a	23.71 c	23.45 d	24.80 a	24.38 b	23.70 c	24.37 b
100-S Weight	Punjab 91	25.79 a	25.84 a	25.16 b	24.58 c	23.47 d	25.43 b	25.13 b	24.66 c	25.00 a
(1)	C 141	26.07 a	26.04 a	25.00 c	24.59 d	24.11 e	25.77 a	25.39 b	24.94cd	25.24 a
	Mean	25.65 a	25.63 a	24.98 c	24.29 d	23.68 e	25.33 b	24.97 c	24.43 d	
	Noor 91	19.60 a	17.66ab	8.47 e	8.84 e	17.03ab	16.37bc	14.79cd	13.31 d	14.45 c
Grain Yield	Punjab 91	35.31 a	32.46 b	23.25 c	20.54 d	12.07 f	16.32 e	14.32 ef	13.34 f	20.95 b
(2)	C 141	23.62 c	22.41 c	17.64 d	21.34 c	20.97 c	27.59 b	33.30 a	18.03 d	23.11 a
	Mean	26.03 a	24.18 b	16.45 d	16.91 d	16.69 d	20.09 c	20.80 c	14.89 e	
	Noor 91	96.54 c	96.03 c	83.95 e	89.23 d	101.36b	105.75a	93.71 c	81.80 f	93.54 b
Biol. Yield	Punjab 91	106.96a	108.45a	90.71 b	105.98a	84.43 c	84.11 c	93.18 b	90.51 b	95.54 b
(3)	C 141	112.93ab	110.20b	101.08c	110.35b	101.03c	115.60a	102.96c	86.88 d	105.13a
	Mean	105.48a	104.89a	91.91 d	101.85b	95.60 c	101.82b	96.62 c	86.40 e	
	Noor 91	19.84 a	18.38ab	10.8 d	9.89 d	16.86bc	15.47 c	15.78 c	16.26 c	15.32 b
Harvest Index	Punjab 91	33.00 a	29.89 b	25.63 c	19.37 d	14.29 e	19.39 d	15.36 e	14.72 e	21.43 a
(4)	C 141	20.92 c	20.34 c	17.46 d	19.36 c	20.76 c	23.86 b	32.33 a	20.75 c	21.97 a
	Mean	24.59 a	22.87 b	17.72 e	16.21 f	17.30 e	19.57 d	21.16 c	17.24 e	
	Noor 91	119.33e	119.33e	124.33c	127.33b	129.33a	120.66e	122.66d	124.66c	123.45b
Flower. Days	Punjab 91	114.33f	114.33f	123.66c	126.66b	128.66a	116.33e	117.66e	119.66d	120.16c
(5)	C 141	119.66e	119.66e	125.33c	128.33b	131.66a	122.33d	123.66cd	124.66c	124.41a
	Mean	117.77g	117.78g	124.44c	127.44b	129.88a	119.77f	121.33e	123.00d	
	Noor 91	166.66ef	166.33f	170.33c	171.66b	177.66a	167.66de	168.00d	170.33c	169.83c
Maturity Days	Punjab 91	169.33c	168.66c	171.66b	175.33a	176.33a	169.33c	169.33c	171.66b	171.50b
(6)	C 141	170.66d	171.00d	174.33b	179.66a	178.66a	171.66d	172.00c	173.33b	173.91a
	Mean	168.88f	168.66f	172.11c	175.55b	177.55a	169.55e	171.00d	170.66d	

	(1)	(2)	(3)	(4)	(5)	(6)
S.x. (V)	0.0392	0.2565	0.2915	0.5597	0.0408	0.0935
S.x. (T)	0.0505	0.3861	0.5405	0.2211	0.2000	0.1825
S.x.(VxT)	0.0875	0.6688	0.9363	0.3829	0.3464	0.3162

Means not followed by the same letter for a character are statistically significant by DMRT at 1.0 % level of probability.

significantly as compared to their respective controls. The irregular response of biological yield with gamma irradiation may be due to the kind and extent of biological damage, while the consistent decrease in biological yield may be accounted for the protective and repairing activity of GA_3. Table 2 showed that in the three genotypes biological yield decreased inconsistently at various gamma irradiation dosages. Post mutagenic application of GA_3 significantly changed the biological effects of gamma irradiation either in positive or negative direction. This suggests that the treatment of GA_3 could be useful for inducing extra variability.

Gamma irradiation decreased the harvest index significantly with both treatments as compared to their respective controls. Contrary to this, Yousaf *et al*. (1991) have recorded little variation in harvest index percentage under different gamma irradiation in lentil. The change in the results might be due to different genotypes and places of experimentation. However, application of GA_3 significantly increased the harvest index percent at 40 and 50 Kr by modulating the effects of gamma irradiation.

Highly significant interaction between genotype and treatment indicate varied response of harvest index towards the various doses of gamma irradiation. Gamma irradiation decreased the harvest index differently at all the treatments in the three genotypes as compared to control. However, with the application of GA_3 stimulation in harvest index over control was recorded in C141 genotype. The results of the present study further reveal that the application of GA_3 has changed the effects of gamma irradiation, which might increase the variability for this character in chickpea.

Gamma irradiation significantly and progressively increased the number of days to 50 percent flowering at various levels of irradiation as compared to control. Late flowering in M_1 generation as compared to control have also been reported in pea (Khan *et al*., 1990; Amjad *et al*., 1993), french bean (Svetleva and Petkova, 1992). However, a non-significant delay in flowering with gamma irradiation was reported by Yousaf *et al*. (1991) in lentil. However, post mutagenic treatment with GA_3 decreased the time to 50 percent flowering.

Highly significant interaction of varieties and doses indicate that the response of varieties to various levels of irradiation is quite variable. The results show a relative delay in 50 percent flowering over control in the three varieties. Application of GA_3 reduced the number of days to 50 percent flowering at different intensities of gamma irradiation. This decrease in time could be due to the repairing process of GA_3, which might bring the population to a physiological state for early flowering.

Gamma rays consistently delayed in crop maturity with an increase of radiation intensity as compared to control. The results obtained in this study are in line with those of Hassan *et al*. (1988) in wheat, Tripathi and Dubey (1992) in lentil and Kumar *et al*. (1993) in pea. Post mutagenic application of GA_3 decreased the time to crop maturity.

A linear increase in time to crop maturity was recorded with gamma irradiation in the three genotypes as compared to control. However, with GA_3 treatment less number of days were taken to maturity at various levels of irradiation in the three genotypes. The application of GA_3 modulated the effects of gamma irradiation either in positive or negative direction. This may leads to the induction of new genetic variation for different characters and widens the germplasm pool. In a breeding programme extent of genetic variability is more important than the total variability. Post mutagenic application of GA_3 may prove to be useful in widening the genetic spectrum and consequently the selection of new genotypes with better genetic architecture in regards with the yield improvement in chickpea.

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