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Effect of Gamma Rays on Germination, Morphology, Yield and Biochemical Studies in Groundnut (*Arachis hypogaea* L.)

A. Gunasekaran[1] and **P. Pavadai**[2,*]

[1]R & D Centre, Bharathiar University, Coimbatore - 641 046, Tami Nadu, India

[2]P.G and Research, Department of Botany, Periyar Government Arts College, Cuddalore - 607 001, Tamil Nadu, India

*E-mail address: drksbotany@gmail.com

ABSTRACT

Groundnut (*Arachis hypogaea* L.) is an important oilseed crop and grain legume grown worldwide. The groundnut seed has dual advantage of being important as a source of edible oil as well as protein. However, it is self pollinating and possesses limited variability. Despite its long history of cultivation, broad sub-specific variability and wide geographic distribution of the cultivated groundnut, defects in its genetic composition with respect to requirement of man are wide spread and for many of these no remedial resources are known to exist among its varietal forms. The exploitation of genetic resources from wild species is extremely difficult because of ploidy differences between cultivated tetraploid and diploid wild species coupled with compatibility barriers except with *Arachis* section. To induce polygenic variability for yield and its components in peanut (*Arachis hypogaea* L.) var. VRI-2 were treated with γ-radiation (10, 20, 30, 40, 50, and 60 KR). Effects of gamma rays treatment were observed in M1 generation gradually reduced in all parameters except days to first flower to increase concentration of treatment. In M_2, M_3 and M_4 populations, the significant increase of grain yields and yield components of Groundnut were observed. Potential high yielding mutants were identified in progenies of treated seeds.

***Keywords*:** Groundnut, mutagens, Oilseeds, Gamma rays, *Arachis hypogaea*

1. INTRODUCTION

Peanut or groundnut (*Arachis hypogaea* L.) is the third largest oilseed crop after soybean and seed cotton, globally. Peanut was cultivated on more than 24 m ha annually during the 2002–04 triennium, producing more than 35 Mt and with productivity of about 1437 kg ha^{-1} peanut-in-shell (FAO 2002–04).As well as producing edible oils, peanut seeds are rich in protein. About two-thirds of world peanut production is used to extract oil and the remainder is in the form of protein-rich, edible products. Peanut is native to southern Bolivia and north-western Argentina. Sixty-nine species are known to occur in the genus *Arachis*, which have been classified into nine sections. Peanut is a tetraploid ($2n = 40$) while some wild species are diploid too. It is a highly self pollinated crop. In many countries, peanut is grown by smallholder farmers as a cash crop. The value of genetically pure and viable germplasm seed for use in crop improvement is well recognized.

The groundnut seed mainly comprised of protein, fat, carbohydrate which make it sensitive to radiation induced stress. Among the environmental stresses, the radiation is the most important factor, which limits production of groundnut. This would result in drastic reduction in crop yield and magnitude of reduction would depend on groundnut varieties. Not only the yield of Groundnut but also the quality of products decrease sunder radiation stress.

The seed stage is a convenient phase in the plant's life cycle for use in radiological studies to determine relative radio sensitivity of species and the effects of various factors on radio sensitivity.

(A)

(B)

(C)

Photo A, B, C. Groundnut (*Arachis hypogaea* L.).

Earlier experiments in this field have indicated that ionizing radiation could cause permanent genetical effects, lethal or beneficial mutations, morphological modifications and other effects in plants.

Several factors may be involved in the inhibition of germination and the growth of the plants from seeds following their exposure to high irradiation doses. Anumber of radiobiological parameters are commonly used in early assessment of effectiveness of radiation. Methods based on physiological changes such as inhibition of seed germination and shoot and root elongation have been reported for detection of irradiated legumes.

Mutation breeding supplement conventional plant breeding as a source of increasing variability and could confer specific improvement without significantly altering its phenotype. The successful utilization of gamma rays to generate genetic variability in plant breeding has been reported in Groundnut (Takagi and Anai) 2006, (Benslimani and Khelifi) 2009 and (Nadaf et al.,) 2009 and other crops. Takagi and Anai in Soybean (2006), Avila and Murty in "Cowpea and Mungbean (1983), Routaray (1995) and Devi and Mullainathan in Blackgram (2012). Therefore, in present study the response of groundnut seed to gamma radiation stress on germination and seedling parameters of ground nut was instigated compared to no irradiated seed.

2. MATERIALS AND METHODS

2. 1. Biological materials

The dry and dormant seeds of the Groundnut (*Arachis hypogia*) var. VRI-2 were obtained from Milled Breeding Station, Tamil Nadu Agricultural University, Coimbatore.

2. 2. Mutagens

Physical mutagens like Gamma rays were used at various doses to induce mutagenesis.

2. 2. 1. Mutagen treatments

Seeds of peanut cultivar VRI-2 were treated with γ-radiation. Uniform size seeds of each cultivar were used for treatment. Treatments (200 seeds per treatment) consisted of six different doses of γ-radiation (10, 20, 30, 40, 50 and 60KR). Untreated seed stock of the respective cultivars was used as a control. Seeds were irradiated with γ-radiation at Sugar cane Breeding Institute, Coimbatore, Tamil Nadu India. Treated seeds were sown in the field plots along with untreated control. The seeds were sown in a randomized complete block design in three replications with spacing of 30 cm between the rows and between plants. The recommended package of practice for the crop was followed.

The M_1 plants were harvested on a single plant basis. M_2 generation seeds were raised from M_1 generation and M_3 and M_4 generation seeds were raised from M_2 and M_3 generation. The seeds were collected from different individual mutagenic treatment. Segregating mutant lines based on visual observations and low performing were discarded in the initial stages of evaluation and progenies were advanced on the basis of superiority of their yield performance over the respective controls, finally ending up with 10 superior mutant lines in M_2 generation. The selected 10 mutant progenies were evaluated in a replicated trial to assess their performance and identify high-yielding mutants.

The 10 mutants, untreated controls (parents) and two checks were grown in randomized complete block design with three replications in a plot of 4.0 m x 2.4 m with spacing of 30 cm x 10 cm over three successive generations. From each entry, 10 plants were randomly selected for recording observations and followed by M_3 and M_4 generation. The data on mean performance to the following quantitative characters, such as days to first flowers, plant height, number of leaves per plant, number of kernal per plant, 100seed weight, kernal yield per plant, oil content, protein content fresh weight and dry weight.

3. FIELD OBSERVATIONS

3. 1. Days to first flower (days)

The number of days taken from sowing to first flower was recorded and expressed as number of days to first flower.

3. 2. Plant height (cm)

The height of the plant from the base to the top of the plant of maturity was measured and expressed in cm.

3. 3. Number of leaves per plant

The number of leaves was counter and recorded as the number of leaves per plants

3. 4. Number of kernal per plant

Total number of kernal at maturity time were counted and recorded as the number of kernal per plant.

3. 5. 100 seed weight

The weight of 100 seed were weight and recorded

3. 6. Kernel yield per plant

Total number of kernel yield from individual plant were counted and recorded as the number of seeds per plant.

3. 7. Protein content

Two seeds from the same plant of each M_1, M_2, M_3 and M_4 generation plants were separately de hulled and ground in a mortar and the extracts were defatted by washing with three changes of cold acetone for 4 to 6 hrs. The acetone was removed by filtration and the extracts were air-dried at room temperature.

The proteins from the defatted meal were precipitated with 10% trichloroacetic acid and recovered by centrifugation at 5000 rpm for 30 minutes at 40 °C. The protein content was then determined calorimetrically according to the method of Lowry *et al*. (1951) using bovine serum albumin as standard.

3. 8. Oil content

The oil content of the seed was estimated with petroleum ether in Soxhlet extraction apparatus (Cox and Pearson, 1962).About 50 g of seed was dried in a drying dish at 130 °C for 20 min. in a forced draft oven. Then they were cooled to room temperature and passed through the nut slicer to slice the nuts. The sliced samples were mixed well and accurately 2 g of the sample was taken in to a filter paper fold. The filter paper was folded in such a way to hold the seed meal. A second filter paper was used to wrap around the seed which was left open at the top like a thimble. The sample packet was placed in the butt tubes of the Soxhlet extraction apparatus. Extraction was done with petroleum ether (150 drops min-1) for 6 hrs without interruption by gentle heating. Then the extraction flask was dismantled after cooling and then ether was evaporated on a water bath until no odor of ether remained. The dirt or moisture outside the flask was carefully removed and the flask was weighed. The heating was repeated to get constant weight

4. RESULTS AND DISCUSSION

The present investigation was undertaken in order to study the induced mutation of the genotype namely Groundnut variety VRI-2 by using Gamma rays (γ). M_2 generation seeds were raised from M_1 generation, the seeds were collected from different individual mutagenic treatment followed for the M_3 and M_4 generation

4. 1. M_1 generation

The M_1 generation results were observed and recorded. The results all parameters such as days to first flowers, plant height, number of branches per plant, number of leaves per plant, number of cluster per plant, number of pod per plant, number of seed per plant, seed yield per plant, decrease with increase dose of mutagenic treatment. The present results confirm these earlier reports in groundnut were observed for gamma rays and NaN3 (Mandal et al., 2007), soybean (Pepol and Pepo, 1989 and Pavadai et al., 2009); mung bean (Khan and Wani 2005) and sesame (Prabhakar 1985).

4. 2. M_2, M_3 and M_4 generation

The data on mean performance to the following quantitative characters, such as days to first flowers, plant height, number of leaves per plant, number of kernal per plant, 100seed weight, kernal yield per plant, oil content, protein content fresh weight and dry weight. In the present study, the mean values recorded a negative and positive shift for the morphology, yield and biochemical content such as such as days to first flowers, plant height, number of leaves per plant, number of kernal per plant, 100seed weight, kernal yield per plant, oil content, protein content fresh weight and dry weight. The maximum yield parameter were recorded at 50 KR gamma rays treatment. Similar effects in groundnut were observed for gamma rays and NaN_3 (Mandal et al., 2007). Similar observations were also made in other plants like black gram (Julite Hepziba and Subramanian, 2002; Arulbalachandran, 2006), Cowpea (Odeigah *et al*., 1998), Okra (Ghai *et al*., 2004) and Sesame (Sengupta and Datta, 2004).soybean (Papa *et al*., 1961; Balakrishnan, 1991; Geetha, 1994; Cheng and Chandlee, 1999; Dhole *et al*., 2003; Pavadai and Dhanavel, 2004 and 2005; Pavadai, 2006).

Table 1. Effect of gamma rays on quantitative parameters of Groundnut in M_1 generation.

Treatment	Control	GAMMA RAYS 10KR	20KR	30KR	40KR	50KR	60KR
Germination %	93.00	86.59	77.92	71.58	61.45	52.36	40.25
Seedling survival %	90.25	84.26	75.50	69.65	58.20	49.45	37.24
Days to first flower (Days)	35.38	36.11	36.20	36.95	37.55	37.94	37.98
Plant height (cm)	42.58	40.05	40.88	41.30	41.80	42.05	42.55
No. of leaves/ plant (30days)	50.12	47.40	46.25	45.85	45.01	40.22	35.85
No. of kernal/ plant	29.50	25.55	22.41	20.13	18.65	16.66	12.54
100 Seed weight (g)	35.30	35.10	35.00	34.85	34.44	32.88	32.11
Kernal yield/ plant (g)	19.25	18.46	17.55	17.11	16.33	15.42	14.46
Fresh weight (g)	95.53	90.13	87.25	83.44	80.26	75.10	70.45
Dry weight (g)	35.28	33.88	31.20	30.05	28.55	26.65	24.82

Table 2. Effect of gamma rays on quantitative parameters of Groundnut in M_2 M_3 and M_4 generation.

Treatment		Days to first flower	Plant height	No. of leaves/ plant (30days)	No. of kernal/ plant	100 Seed weight (g)	Kernal yield/ Plant (g)	Oil contend	Protein contend	Fresh weight (g)
M_2 Generation	Control	36.28	43.87	48.63	28.76	30.27	25.64	49.12	34.58	85.36
	GAMMA RAYS 10KR	35.14	44.15	45.24	25.38	28.65	26.41	47.69	33.21	82.40
	20KR	35.07	47.65	46.32	26.54	28.14	28.16	46.88	33.84	88.59
	30KR	35.98	50.11	50.14	29.22	30.45	27.44	50.13	34.66	84.51
	40KR	34.88	50.48	52.87	30.14	31.25	28.98	50.58	35.21	90.04
	50KR	34.32	52.89	55.35	32.87	33.69	30.15	50.88	35.86	90.63
	60KR	33.88	51.25	50.47	27.45	30.14	25.54	48.10	34.26	82.51
M_3 Generation	Control	35.21	45.46	45.62	25.30	29.88	23.28	48.72	33.32	83.81
	GAMMA RAYS 10KR	34.56	45.25	44.32	24.85	28.59	24.12	47.81	32.25	84.36
	20KR	34.42	46.11	45.16	24.15	29.36	24.52	47.54	32.56	84.65
	30KR	33.85	46.54	45.84	25.64	30.11	24.85	48.13	33.65	85.07
	40KR	33.32	45.47	46.35	26.50	30.24	25.36	48.21	33.81	85.65
	50KR	33.54	46.87	46.88	28.17	30.69	27.11	48.53	34.18	86.33
	60KR	34.18	43.55	42.08	23.61	30.48	23.29	47.44	34.66	85.10
M_4 Generation	Control	34.32	42.19	43.77	27.28	30.04	24.41	49.05	34.06	84.05
	GAMMA RAYS 10KR	34.10	41.36	41.29	27.64	29.85	23.65	48.75	34.12	84.57
	20KR	33.29	42.05	40.47	26.53	29.61	24.58	48.56	34.25	85.36

	30KR	33.56	42.77	44.54	27.99	30.18	25.13	48.82	34.80	85.20
	40KR	33.05	42.95	44.70	29.48	31.04	25.33	49.30	35.14	85.99
	50KR	32.85	43.22	45.31	29.56	31.21	26.98	49.81	35.62	86.23
	60KR	33.62	40.13	43.44	23.05	30.10	25.41	47.23	35.21	83.29

5. CONCLUSION

In M_2, M_3 and M_4 generation all parameters were recorded in moderate and high value. The highest mean value for all parameters was recorded in 50 KR gamma rays treatment than the other treatments with control.

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