

## Effect of Mutagens on Seed Quality Characters in Groundnut (*Arachis hypogaea* L)

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### ABSTRACT

An experiment was conducted during 2012-13 to study polygenic variability on seed quality traits in groundnut (*Arachis hypogaea* L.) through mutagens. Groundnut genotype CTMG-6-1 was treated with gamma radiation (20, 30 and 40 kR) and ethyl methane sulphonate (EMS- 40mM and 60mM) for studying seed germination, seedling length, seedling dry weight, seedling vigour index (SVI) and electrical conductivity. It was observed that the germination percentage, speed of germination index, seedling length and seedling vigour index decreases with increasing the doses and concentrations of gamma rays and EMS in M<sub>1</sub> and M<sub>2</sub> generations when compared to control. The LD50 (Lethal dose) value was determined based upon the seed germination percentage. Seedling length and seedling vigour index observed higher in M<sub>2</sub> generation as compared to M<sub>1</sub> generations. Also revealed that the increase in the doses of mutagenic treatments of gamma rays and EMS, electrical conductivity of seeds were increased.

**Key words** EMS, Gamma ray, groundnut, seed quality

Groundnut (*Arachis hypogaea* L.) is grown in majority of countries of the world and plays an important role in world economy. It is known by several vernaculars as peanut, monkey-nut or goober nut etc. (Reddy, 1988). It is the single largest source of edible oils and constitutes roughly about 50 per cent of the total oil seeds production. Artificially induced mutation is one of method to enhance genetic variability within short time. Recently, peanut has been gaining importance as a food crop, due to its high content of digestible proteins, vitamins, minerals, and phytosterols, and to increased consumer preference after value addition (Nadaf *et.al.* 2009). Sustained induced mutagenesis in oilseeds using Gamma-rays and ethyl

methane sulphonate resulted in wide spectrum of mutants affecting various traits. Planned irradiation had broken undesirable linkages and enhanced favorable recombinants. As a result, traits like large seed, increased harvest index, assimilate partitioning, semi-dwarf habit, earliness, new ideotypes, improved seed quality, and enhanced disease resistance were incorporated in oilseed (Souza *et al.*, 2009). Several mutant genes have been successfully introduced into commercial crop varieties that significantly enhance the nutritional value. (Jain and Suprasanna, 2011). The main advantage of mutation breeding is the possibility of improving one or two characters without changing the rest of the genotype (Lukanda *et al.*, 2013). Gamma rays are the most energetic form of electromagnetic radiation, their energy level is from ten to several hundred kilo electron volts and they are considered as the most penetrating compared to other radiations (Kovacs and Keresztes 2002). Gamma radiation can be useful for the alteration of physiological characters (Kiong *et al.*, 2008). Ethyl Methane Sulphonate (EMS) is mutagenic and carcinogenic organic compound, it produces random mutations in genetic material by nucleotide substitution; particularly by guanine alkylation and it is reported to be the most effective and powerful mutagen (Hajara, 1979) and typically produces only point mutations (Okagaki *et al.*, 1991).

### MATERIALS AND METHODS

The experimental material consists effect of mutagenesis on groundnut genotype CTMG-6-1. Seeds of groundnut genotype were treated with gamma radiation and ethyl methane sulphonate (EMS). Uniform size seeds of each cultivar were used for treatment. Treatments (200 seeds per treatment) consisted of three different doses of gamma radiation (20, 30 and 40 kR) and two doses

**Table 1. Analysis of variance for different seed quality traits of different mutagenic treated groundnut in M<sub>1</sub> generation.**

Sources of Variation	d. f.	GP (%)	SGI	EC (dSm <sup>-1</sup> )	MC (%)	SDW (g)	SSL (cm)	SRL (cm)	SL (cm)	SVI
Treatment	5	156.96**	70.07**	0.13**	0.16*	6.87 E-03**	10.84**	26.75**	64.36*	671688.80**
Error	18	13.72	2.70	5.62 E-04	0.052	5.61 E-04	0.085	0.13	0.31	8755.11

\*Significant at 5 %, \*\*Significant at 1 %

of EMS (40 and 60 mM). Untreated seed stock of the respective genotype was used as a control. Seeds were irradiated with  $\gamma$ -radiation at Bhabha Atomic Research Center (BARC) Mumbai, India. EMS solution was prepared in 0.1 M phosphate buffer (pH = 7.0). Seeds were presoaked in distilled water for eight hours to allow uptake of ethyl methane sulphonate (EMS). Presoaked seeds were treated with EMS for two hours at room temperature in cloth bags. Treated seeds were then rinsed in running tap water for four hours and sown in the field plots along with untreated control. Seeds were treated with EMS at Biotechnology Centre, Dr. PDKV, Akola. The effect of gamma rays and EMS treatments was studied with respect to the germination percentage; seedling length, seedling vigour index, and electric conductivity were analyzed in laboratory condition. The treated genotype was subjected to laboratory experiments

by as per procedure of Completely Randomized Design illustrated by Rangaswami (1995) for determination the effects of the physical and chemical mutagen on seed quality traits for M<sub>1</sub> and M<sub>2</sub> generations.

## RESULTS AND DISCUSSION

Induced mutagenesis due to various mutagenic treatments was measured in terms of mean in M<sub>1</sub> and M<sub>2</sub> for seed quality traits. The effect of gamma rays and EMS treatments on germination percentage, speed of germination index, seedling length, seedling vigour index and electrical conductivity were given in Table 3. Also Table 3, revealed that the maximum germination percentage of (86.00) per cent was recorded in 40 mM followed by 60 mM (82.00%), which were statistically superior to that of control (74.00%). However, germination percentage was adversely

**Table 2. Analysis of variance for different seed quality traits of different mutagenic treated groundnut in M<sub>2</sub> generation.**

Sources of Variation	d.f.	GP (%)	SGI	EC (dSm <sup>-1</sup> )	MC (%)	SDW (g)	SSL (cm)	SRL (cm)	SL (cm)	SVI
Treatment	5	87.46**	28.13**	3.55 E-02**	0.11**	2.39 E-03**	2.29**	7.13**	12.30**	252860.80**
Error	18	3.55	0.22	6.98 E-04	0.027	3.63 E-04	0.52	1.06	1.01	18375.11

\*Significant at 5 %, \*\*Significant at 1 %

d.f.- Degrees of freedom

GP-Germination percentage, SGI- Speed of germination index, EC-Electrical conductivity, MC-Moisture content SDW-Seedling dry weight SSL-Seedling Shoot Length (cm), SRL-Seedling Root Length(cm) , SL- Seedling length (cm), SVI-Seedling Vigour Index

**Table 3. Effect of gamma rays and EMS on seed quality traits in M<sub>1</sub> and M<sub>2</sub> generations**

Treatments	Germination percentage (%)		Speed of germination index (%)		Electrical conductivity		Moisture content (%)		Seedling Dry Weight (cm)		Seedling shoot length (cm)	
	M <sub>1</sub>	M <sub>2</sub>	M <sub>1</sub>	M <sub>2</sub>	M <sub>1</sub>	M <sub>2</sub>	M <sub>1</sub>	M <sub>2</sub>	M <sub>1</sub>	M <sub>2</sub>	M <sub>1</sub>	M <sub>2</sub>
20 kR Gamma	76.00	91.25	14.11	26.32	0.88	0.30	7.3	6.2	0.33	0.33	7.50	12.75
30 kR Gamma	69.50	85.75	12.76	23.33	0.92	0.33	7.5	6.2	0.31	0.30	5.70	12.41
40 kR Gamma	72.00	81.25	12.19	22.73	0.98	0.52	7.3	6.5	0.33	0.27	5.45	10.72
40 mM EMS	86.00	89.50	22.31	21.65	0.54	0.45	6.9	6.3	0.43	0.33	9.82	11.76
60 mM EMS	82.00	79.12	19.70	19.18	0.59	0.27	7.1	6.5	0.39	0.31	6.62	11.67
Control	74.00	82.00	13.60	19.50	0.81	0.41	7.2	6.1	0.36	0.32	5.87	11.14
F test	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.
SE(m)±	1.85	0.94	0.82	0.23	0.011	0.013	0.11	0.08	0.011	0.0095	0.14	0.36
CD at 5 %	5.50	2.80	2.44	0.69	0.035	0.039	0.33	0.24	0.035	0.028	0.43	1.07

Table 3 cont... Effect of gamma rays and EMS on seed quality traits in M<sub>1</sub> and M<sub>2</sub> generations

Treatments	Seedling Root Length (cm)		Seedling Length (cm)		Seedling Vigour Index	
	M <sub>1</sub>	M <sub>2</sub>	M <sub>1</sub>	M <sub>2</sub>	M <sub>1</sub>	M <sub>2</sub>
20 kR Gamma	14.37	16.95	21.87	29.71	1663.25	2709.58
30 kR Gamma	8.60	14.62	14.30	27.03	994.92	2348.32
40 kR Gamma	8.20	14.51	13.65	25.24	984.00	2063.30
40 mM EMS	13.62	17.61	23.45	29.37	2017.50	2627.98
60 mM EMS	11.95	17.06	18.57	28.74	1523.67	2291.48
Control	10.45	15.55	16.32	26.69	1208.75	2186.93
F test	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.
SE(m)±	0.18	0.51	0.27	0.50	46.78	67.77
CD at 5 %	0.54	1.53	0.82	1.49	138.94	201.28

affected by doses of gamma rays. In M<sub>2</sub> generation, it was observed that the germination percentage decreases with increasing the doses of mutagenic agents. Maximum germination percentage was observed in 20 kR (91.25%) followed by 40 kR (89.50%) and 30 kR (85.75%) which showed the statistical significance over control (82.00 %). The lowest germination percentage was observed in 60 mM (79.12). The reduction might be due to the disturbance caused at physiological level at cellular

level or acute chromosomal damage (Sinha and Godward, 1973). The germination percentage decreased with increasing dose of gamma rays reported by Singh *et al.* (1988), Badigannawar and Murthy (2007), Jagadeesan *et al.* (2008), Lukanda *et al.* (2012), Aparna *et al.* (2013) in groundnut, Khan and Sonu (2009) in mungbean. Mahla *et al.* (2010) reported that the gradual reduction in germination of the treated population with increased dose of gamma rays and EMS in clusterbean.

Muralidharan and Rajendran (2011) in okra, Roychowdhari and Toh (2011) in *Dianthus* spp. reported due to EMS, Bhosale and Hallale (2011) in blackgram, Dehappour *et al.* (2011) in rice (*Oryza sativa*). The speed of germination index, In M1 and M2 generations speed of germination index indicates the progressive performance of the seed for germination over period. Speed of germination index provides good seed vigour which facilitate in categorizing strong and weak seedling and represent potential of seedling for successful establishment. However, the maximum speed of germination index recorded in treatment 40 mM EMS in M1 generation. While the lowest magnitude of speed of germination index was recorded in 40 kR gamma rays in M1 generation. In M2 generation, the maximum value for speed of germination index recorded in 20 kR (26.32) followed by 30 kR (23.33) and 40 kR (22.73) as compared to control (19.50). Krishnaswamy and Seshu (1989) opined that the rate of germination was positively correlated with oxygen uptake, dehydrogenase activity by providing energy to the germinating embryo and interfering with integrity and overall capacity of the metabolic machinery of the young germinating primordia. Speed of germination index decreases with increased doses of gamma rays. Similar results reported by Aparna *et al.* (2013) in groundnut. From the Table 3, it revealed that the increase in the doses of mutagenic treatments of gamma rays and EMS, electrical conductivity was increased. As per the studies the electrical conductivity was negative related to seed quality aspects in M1 generation, the treatments of gamma rays shown higher magnitude of for electrical conductivity whereas, EMS treatment shows lower values of electrical conductivity than the control (0.81 dSm<sup>-1</sup>). In M2 generation all the treatments shown lower magnitude than M1 generation for each treatment. Electrical conductivity of the seeds exposed to gamma rays higher than that of unirradiated seed (control) finding by Amjad and Anjum (2002) in onion. From table 3, The results revealed that the moisture content in M<sub>1</sub> generation was higher than those of M<sub>2</sub> generation for all treatments. Table 3, showed that in M1 generation, the maximum seedling dry weight recorded in 40 mM (0.43 g) EMS while lowest seedling dry weight in 20 kR (0.33g) and 40 kR (0.33 g) gamma rays over its control (0.36 g). In M2 generation, the maximum

seedling dry weight of 0.33g recorded in 20 kR and 40 kR. It can be concluded that the increase in the doses of gamma rays and EMS decreases the seedling dry weight. Increase in the doses of gamma rays and EMS, seedling dry weight was decreased concluded negative relation with increase mutagenic doses. Borzouei *et al.* (2010) reported that seedling dry weight decreased with increasing radiation doses in wheat. Table 3, showed that in M1 generation decrease shoot length was observed with increase in doses of mutagens viz. gamma rays and EMS respectively. Seedling shoot length was decreased with increasing concentration and doses of and gamma rays and EMS respectively. Shakoor (1978) and Khalil (1986) attributed decreased shoot length at higher doses of gamma rays to reduced mitotic activity in meristamatic tissues and reduced moisture contents in seeds respectively. Similar results found by Lukanda *et al.* (2012) and Aparna *et al.* (2013) in groundnut. Borzouei *et al.* (2010) in wheat, Muralidharan and Rajendran (2011) in Okra, Talebi *et al.* (2012) in paddy, Dehappour *et al.* (2011) in rice (*Oryza sativa*). Table 3, indicated the decrease root length was observed with increase dose of both mutagens gamma rays and EMS in M1 and M2 generations. Seedling root length was decreased with increasing concentration and doses of both gamma rays and EMS respectively. Shakoor (1978) and Khalil (1986) attributed decreased root length at higher doses of gamma rays may results to reduced mitotic activity in meristamatic tissues and reduced moisture contents in seeds respectively. Similar results found by Lukanda *et al.* (2012) and Aparna *et al.* (2013) in groundnut, Borzouei *et al.* (2010) in wheat, Muralidharan and Rajendran (2011) in okra, Talebi *et al.* (2012) in paddy. Seedling length was decreased with increasing concentration and doses of EMS and gamma rays respectively. Kiong (2008) reported that radiation increases plant sensitivity to gamma rays and this may be caused by the reduced amount of endogenous growth regulators, especially cytokinins, as a result of break down, or lack of synthesis, due to radiation and noticed that treating seeds with high rates of gamma radiation reduced germination with a corresponding decline in growth of plants. Similar results found by Lukanda *et al.* (2012) and Aparna *et al.* (2013) in groundnut, Borzouei *et al.* (2010) in wheat, Muralidharan and Rajendran (2011) in okra, Talebi

*et al.* (2012) in paddy. The range for seedling vigour index in M1 generation was observed between 984.00 to 2017.50 and 2063.00 to 2709.58 in M2 generation. Seedling vigour index decreases with increasing the dose of gamma rays and EMS. The gradual reduction in root and shoot length with increase in gamma dose also resulted in corresponding decrease in seedling vigour index from 20 kR (1663.25) due to 40 kR (984.00) of gamma rays. Seedling vigour index decreased with increase the doses and concentration of gamma rays and EMS. It indicates the relative sensitivity of groundnut for varying dose of mutagenic treatments include radiation (Gamma rays) and chemical (EMS) agents resulting on overall vigour of seedling and subsequent establishment of mutants. Similar results reported by Aparna *et al.* (2013) in groundnut, Muralidharan and Rajendran (2011) in okra. Seed germination percentage and seedling traits was inhibited due to increasing doses/concentrations of mutagens. Almost all the mutagenic treatments caused decrease in seedling length and seedling vigour index in a groundnut.

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