



Studies on mutagenic effectiveness and efficiency of finger millet [*Eleucina coracana* (L.) Gaertn] in M₁ generation and effect of gamma rays on its quantitative traits during M₂ generation

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Abstract : Dry seeds (12 % moisture) of two finger millet cultivar viz., Dapoli-1 and Dapoli Safed were irradiated with four doses of gamma-rays viz., 400 Gy, 500 Gy, 600 Gy and 700 Gy at BARC, Mumbai. In laboratory test, root and shoot lengths of seedlings were decreased with increase in dose of gamma rays. Similarly, germination percentage and survival rate of seedlings were decreased with increase in dose of gamma irradiation during field study. In M₁ generation, three types of chlorophyll mutations viz., *albino*, *xantha* and *viridis* were observed. *Albino* and *xantha* were observed in all treatments, whereas, *viridis* observed only in lower doses viz., 400 Gy and 500 Gy. Based on the chlorophyll mutation frequency on M₁ plants, mutagenic effectiveness and efficiency were computed. In Dapoli-1 variety, two early maturing mutants and three high yielding mutants were isolated from 500 Gy dose and 600 Gy dose, respectively. In M₂ generation the mutagenic treatments were effective in inducing various types of chlorophyll and morphological macro mutants, few of those showed significant change in flowering, maturity and plant height characters and few of them have good breeding value.

Key Words : Finger millet, Chlorophyll mutations, Macro mutants, Gamma rays

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INTRODUCTION

Among the major food grains, finger millet [*Eleucina coracana* (L.) Gaertn] is one of the most nutritious crops. It is an important food crop in South Asia and Africa. The grain of finger millet has fine aroma when cooked or roasted and it is known to have many healthy promising qualities. It is a rich source of calcium and has good amount of magnesium, phosphorus and iron. Finger millet has a favorable amino acid spectrum that includes cysteine, tyrosine, tryptophan and

methionine (Rachie, 1975). Genetic improvement of crop depends on the amount of genetic variability present in the population.

Mutation breeding is the tool in the hand of breeder to create variability in crop population and to make selection in the population with the view to bring about further improvement in crop. Mutations are theoretical changes which occur in DNA sequences and results in change in genetic code. A gene mutation or point mutation is the group of all heritable changes which occur within the limit of a single gene.

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In general mutation breeding has been playing a key role in self-pollinated crop with limited variability. Mutation breeding has been reported by many workers, in castor (Ankineedu *et al.*, 1968), in wheat (Swaminathan, 1969), in sesame (Sharma, 1993), in cowpea (Dhanavel *et al.*, 2008), in black gram (Thilagavathi and Mullainathan, 2009) and soybean (Padmavathi *et al.*, 1992 and Pavadai *et al.*, 2010) developed plant by mutation breeding. Gamma irradiation as mutagen can induce useful as well as harmful mutation in plants (Gupta, 1996; Micke and Domini, 1993). The present investigation was undertaken to study the mutagenic effectiveness and efficiency in M_1 generation and to study effect of gamma rays in quantitative characters of finger millet in M_2 generation and results are discussed.

MATERIAL AND METHODS

The material for this study comprised of two varieties of finger millet, Dapoli-1 (mid-tall, fully open ear heads with brown seeds) and Dapoli Safed (mid-tall, partially open ear heads with white colour seeds). Dry seeds of both varieties were irradiated with four doses of gamma rays (^{60}Co) viz., 400 Gy, 500 Gy, 600 Gy and 700Gy at BARC, Mumbai. The experiments to determine the effect of gamma-rays on germination, root and shoot length were conducted on germination paper. Each treatment was replicated five times and for each replication one hundred seeds were sown and tested for their germination, survival, root and shoot length. For determination of LD_{50} observations on germination were recorded on seventh day from the date of sowing. Effect on root and shoot was measured in terms of length of root and shoot, respectively on seventh day. Ten seedlings were selected randomly for taking observations. Chlorophyll mutants were scored and classified. Desirable mutants from M_1 generation were selected on the basis of their phenotypical characters and harvested separately.

Seed harvested from individual M_1 plants were grown as M_2 generation in Factorial Randomized Block Design (FRBD). Two hundred forty seedlings were sown in each plot at spacing of 10×30 cm as well as isolated mutants were also grown individually. All recommended package of practices were followed during growth period of the crop. Newly evolved characters were recorded in M_2 generation. Observation on days to 50% flowering and maturity duration was recorded on plot basis. Observations were recorded on 5 randomly selected plants from each plot of each treatment. In M_2 generation, chlorophyll and morphological macromutants were identified and harvested separately. Mutation frequency was calculated as percentage of M_2 plants and mutagenic effectiveness and efficiency were calculated on the basis of formula suggested by Konzak *et al.* (1965).

$$\text{Mutagenic effectiveness} = \frac{M}{\text{Dose of mutagen (krad)}}$$

$$\text{Mutagenic efficiency} = \frac{M}{L} \text{ or } \frac{M}{I} \text{ or } \frac{M}{S}$$

where,

M = Frequency expressed as percentage of chlorophyll mutation in M_2 generation, estimated on M_2 plant basis.

krad = Kilorad

L = Percentage of lethality of reduction in survival.

I = Percentage of injury or reduction in seedling height.

S = Percentage of panicle sterility.

The data of all characters were statistically analyzed with statistical analysis system software (SAS) V. 9.1 (June 2006), SAS Institute.

RESULTS AND DISCUSSION

Noticeable variations were observed in germination percentage after gamma irradiation in paper germination test. But variation was neither proportional to the increase in dosages nor definite pattern was found in both the varieties studied. On field level germination percentage decreased with increase in dose of gamma rays in both varieties (Fig. 1). Similar result have were reported by Ando (1970) in rice, Pathak and Patel (1988), Singh *et al.* (1998), Cheema and Atta (2003), Harding *et al.* (2012) and Talebi and Talebi (2012).

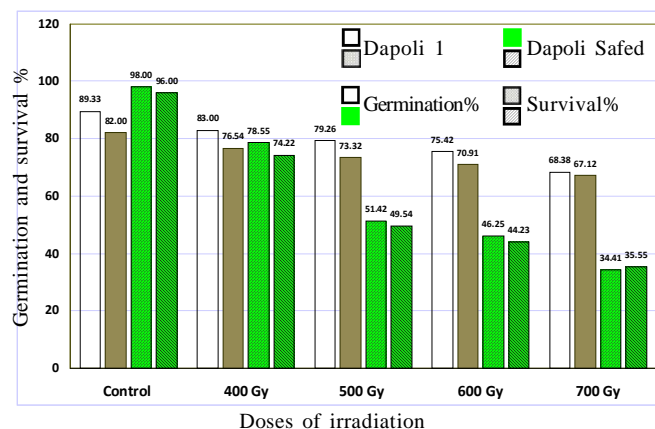


Fig. 1 : Germination and survival percentage affected by gamma rays irradiation in two varieties of finger millet (M_1 generation)

LD_{50} was optimized based on reduction in root and shoot length. It was observed that at 500 Gy had 50 % reduction in root and shoot growth as compared to control (Table1). It was also observed that the root and shoot length decreased with increase in gamma rays dose on approximately linear mode. Similar result was reported by Talebi and Talebi (2012) in rice.

Among the treatments, the 600 Gy and 700 Gy gamma rays produced high frequency of *albino* (Table 1). Next common chlorophyll mutant observed was *xantha* in both varieties. *Viridis* mutants were less frequent and found only in certain treatments viz., 400 Gy and 500 Gy; the frequency of

chlorophyll mutations varied with the genotype as well as mutagen doses in M_1 generation. Total frequency of chlorophyll mutations was relatively higher in Dapoli-1 than Dapoli Safed. The differential response of genotypes to induction of chlorophyll mutations was possible due to differences in the genetic makeup of the varieties used for mutagenesis. During the present study, *albino* mutant occurred in higher frequency than *xantha* or *viridis*. Several workers also reported a higher frequency of albino mutant in irradiated population, (Ando (1970), Singh *et al.* (1998), Cheema and Atta (2003), Chakravarti *et al.* (2013) in rice;

Subramanian *et al.* (2011) in kodo millet).

Mutagenic effectiveness (mutations per unit dose) varied with doses in different genotypes given in Table 2. It was also noticed that, mutagenic effectiveness decreased with increase in strength of gamma rays in both the genotypes, indicating that positive relationship between effectiveness and dose of mutagen. Similar observations of general decrease in effectiveness with increasing doses of gamma rays irradiation was reported in finger millet by Muduli and Misra (2007), in Mung bean by Solanki and Sharma (1994) and in foxtail millet by Gupta and Yashvir (1975).

Table 1 :Root and shoot length and spectrum of chlorophyll mutation

Doses	Variety	Root length (cm)	Shoot length (cm)	Mutant seedlings	<i>Albino</i>	<i>Xantha</i>	<i>Viridis</i>
Control	Dapoli-1	5.0	4.64	–	–	–	–
	Dapoli Safed	7.06	3.75	–	–	–	–
400 Gy	Dapoli-1	3.8 (24.00)	3.10 (33.19)	27	55.55	33.33	11.11
	Dapoli Safed	4.75 (32.72)	2.43 (35.20)	21	57.14	33.33	9.52
500 Gy	Dapoli-1	2.6 (48.00)	2.25 (51.51)	21	52.38	33.33	14.28
	Dapoli Safed	3.58 (49.29)	1.95 (48.00)	19	78.94	15.78	2.26
600 Gy	Dapoli-1	1.35 (73.00)	1.60 (65.52)	18	72.22	27.77	–
	Dapoli Safed	3.45 (51.33)	1.95 (48.00)	17	76.47	23.52	–
700 Gy	Dapoli-1	1.2 (76.00)	1.45 (68.75)	16	87.50	25.00	–
	Dapoli Safed	3.25 (53.96)	1.48 (60.53)	15	80.00	20.00	–

Figures in parenthesis indicate per cent reduction

Table 2: Mutagenic effectiveness and efficiency in M_1 generation

Doses	Mutagenic effectiveness		Mutagenic efficiency	
	Dapoli-1	Dapoli Safed	Dapoli-1	Dapoli Safed
Control	–	–	–	–
400 Gy	0.675	0.525	1.13	1.10
500 Gy	0.420	0.380	1.16	1.05
600 Gy	0.300	0.283	1.00	1.00
700 Gy	0.283	0.214	1.00	1.00

Table 3: Mean values of quantitative characters in M_2 generation of variety Dapoli-1 and Dapoli Safed

Sr. No	Character	Control		400 Gy		500 Gy		600 Gy		700 Gy		Sem ±		C.D. (P=0.05)	
		DPL1	DS	DPL1	DS	DPL1	DS	DPL1	DS	DPL1	DS	DPL1	DS	DPL1	DS
1.	Days to first flowering	93.66	99.54	100.50	105.20	105.21	108.91	106.40	113.74	115.57	120.57	0.19	0.60	0.75	2.37
2.	Days to first panicle maturity	127	135.42	135.72	143.42	138.04	150.32	145.01	152.24	150.13	160.13	0.85	0.44	3.36	1.76
3.	Plant height (cm)	36.28	36.39	30.07	34.05	30.67	33.45	29.66	33.25	33.13	28.57	0.95	1.44	3.74	5.66
4.	Tillers/plant	1.85	1.56	4.48	1.94	4.53	2.86	6.58	3.95	3.05	2.7	0.21	0.43	0.82	1.68
5.	Panicles/plant	1.15	1.06	4.08	3.35	4.05	2.63	5.76	2.60	3.26	2.43	0.61	0.29	2.38	1.13
6.	Fingers/panicle	6.35	6.66	8.40	8.06	8.93	9.13	7.53	9.05	8.66	5.68	0.39	0.65	1.54	2.54
7.	Finger length (cm)	5.77	5.4	8.16	7.02	7.91	7.59	8.23	6.15	7.70	5.94	0.41	0.44	1.61	1.75
8.	Panicle weight/ plant (g)	1.84	1.56	1.93	2.12	1.91	2.86	2.81	2.56	2.25	1.90	0.47	0.17	1.86	0.65
9.	Grain density/cm	46.66	35.83	44.95	38.23	48.9	47.80	57.95	45.33	45.15	35.7	3.58	4.75	14.06	18.66
10.	Yield per plant (g)	3.121	3.926	4.2526	3.3606	4.7494	4.8096	5.1817	4.3951	4.2972	4.115	0.24	0.07	0.94	0.27

DPL1- Dapoli 1

DS- Dapoli Safed

Mean value for quantitative characters are presented in Table 3. In most of the treatments, average increase in number of days to first flowering was observed than control in both varieties. The maximum increase in number of days was seen at 700 Gy (120 days) in Dapoli safed, while minimum increase in number of days for first flowering at 400 Gy in both the varieties than compared to untreated seed. But two early mutants (85 and 86 days to first flowering) were also observed in Dapoli 1 variety at 600 Gy dose of gamma irradiation. This result is in agreement with results obtained by Nirmalkumari *et al.* (2007) in little millet (*Panicum sumatrense*). The maximum mean value for days to maturity was recorded at 700 Gy dose in both cultivar. Significant difference was observed in plant height at 400 Gy, 500 Gy and 600 Gy for Dapoli 1 whereas in Dapoli safed cultivar significant difference was observed at 700 Gy dose of gamma rays. The maximum number of tillers at 600 Gy dose of gamma rays (6.58 and 3.95) in both cultivar as compared to control (1.85 and 1.56). All the mutagenic treatments increased the number of tillers when compared with control. The number of panicles were increased at all doses of gamma rays treatment. Significantly superior difference was observed in all doses of gamma rays treatments for fingers panicle⁻¹ as compared to Dapoli 1 and Dapoli Safed. The maximum fingers were observed at 500 Gy (9.13 and 8.93) plant⁻¹ for Dapoli Safed and Dapoli 1, respectively. The maximum finger length (8.23 cm) was observed at 600 Gy in Dapoli 1, whereas, Dapoli Safed recorded maximum finger length (7.59 cm) at 500 Gy. Similar results were also reported by Muduli and Misra (2008) in finger millet. It indicates that effect of doses of gamma rays varied from genotype to genotype. All the mutagenic treatments showed increase in weight of panicle plant⁻¹ as compared to control. The maximum weight of panicle (2.81 g) was recorded at 600 Gy in Dapoli 1, whereas Dapoli Safed recorded maximum weight (2.86 g) at 500 Gy.

The macro mutations were recorded in grain density cm⁻¹ and yield plant⁻¹ in both varieties of finger millet. The maximum grain density (57.95 grains cm⁻¹) was observed in Dapoli 1 at 600 Gy, whereas Dapoli Safed recorded maximum grain density (47.80 grains cm⁻¹) at 500 Gy. The maximum grain yield plant⁻¹ (5.18 g) was observed at 600 Gy in Dapoli 1 and 4.80 g in Dapoli Safed at 500 Gy. The increase in grain yield plant⁻¹ is inversely related to intensity and radiation doses. These results are in agreement with Chakravarti *et al.* (2013) in rice and Hayat *et al.* (1990) in sorghum cultivars. A number of morphological mutations have been reported in cereal plants and several of these mutations have been shown to exhibit modification in more than one character.

Conclusion :

The improved varieties of finger millet *i.e.* Dapoli-1 and Dapoli Safed showed positive relationship between mutagenic effectiveness and efficiency. The cultivar Dapoli-1 and Dapoli Safed responded more and more number of viable and

economic mutants for higher productivity at 500 Gy and 600 Gy, respectively than other mutagenic treatments. The present investigation revealed that the isolation of early maturity with high yield and yield component characters is possible in 500 Gy and 600 Gy doses of gamma irradiation in finger millet.

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REFERENCES

- Ando, A. (1970). Mutation induction in rice by radiation combined with chemical protectants and mutagens. Rice Breeding with Induced Mutations II, *IAEA bulletin*, Vienna, pp. 1-5.
- Ankineedu, G., Sharma, K.D. and Kulkarni, L.G. (1968). Effects of fast neutrons and gamma rays on castor. *Indian J. Genet.*, **28** : 37-39.
- Chakravarti, K.R., Singh, S., Kumar, H., Lal, J.P. and Vishwakarma, M.K. (2013). Study of induced polygenic variability in M₁ and chlorophyll mutations in M₂ generations in aromatic rice. *Bioscan*, **8**(1) : 49-53.
- Cheema, A.A. and Atta, B.M. (2003). Radiosensitivity studies in basmati rice. *Pak. J. Bot.*, **35**(2) : 197-207.
- Dhanavel, D., Pavadai, P., Mullainathan, L., Mohana, D., Raju, G., Girija, M. and Thilagavathi, C. (2008). Effectiveness and efficiency of chemical mutagens in cowpea [*Vigna unguiculata* (L.) Walp.]. *Afr. J. Biotech.*, **7** : 4116-4117.
- Gupta, P.K. and Yashvir (1975). Induced mutations in foxtail millet (*Setaria italica* Beauv.). Chlorophyll mutations induced by gamma rays, EMS and DES. *Theor. & Appl. Genet.*, **45**(6) : 242-249.
- Gupta, P.K. (1996). Mutation breeding in mungbean, In: A.N. Asthana and D.H.Kim (Eds.) *Recent advances in mungbean research*, pp. 124-136.
- Harding, S.S., Johnson, S.D., Taylor, D.R., Dixon, C.A. and Turay, M.Y. (2012). Effect of gamma rays on seed germination, seedling height, survival percentage and tiller production in some rice varieties cultivated in Sierra Leone. *American J. Exp. Agric.*, **2**(2) : 247-255.
- Hayat, K., Khan, A., Sadiq, M., Elahi, F. and Shakoor, A. (1990). Gamma radiation induced variation in sorghum cultivars. *Pakistan J. Agric. Res.*, **11**(1) : 13-16.
- Konzak, C.P., Wagner, R.A., Nilan, J. and Foster, R.J. (1965). Efficient chemical mutagenesis. *Radiat. Bot. (Suppl.)*, **5**:49-70.
- Micke, A. and Domini, B. (1993). Induced mutations. In Hyward MD, Bosemark NO and Ramagosa I (Eds.) *Plant breeding principles and prospects*. Chapman and Hall, London, pp.52-62.
- Muduli, K.C. and Misra, R.C. (2007). Efficacy of mutagenic treatments in producing useful mutants in finger millet (*Eleusine coracana* Gaertn.) *Indian J. Genet.*, **67**(3) : 232-237.
- Muduli, K.C. and Misra, R.C. (2008). Induced polygenic variability in M₂ generation and its relationship with production of high-yielding mutants in finger millet. *Indian J. Genet.*, **68**(4) : 419-425.

- Nirmalakumari, A., Arulselvi, S., Ganapathy, S., Souframanian, J., Senthil, N. and Devan, P. (2007).** Gamma ray induced variation for lodging resistance and its associated characters in little millet (*Panicum sumatrense* Roth Ex-roem and schult). *Madras Agric. J.*, **94**(7-12) : 151-155.
- Padmavathi, T., Devi, P. and Kiranmai, V. (1992).** Induced variability for different biological parameters in soybean. *J. Cytol. Genet.*, **27** : 175-177.
- Pathak, H.C. and Patel, M.S. (1988).** Sensitivity of upland rice genotypes to gamma radiation. *IRRN*, **13** : 6-9.
- Pavadai, P., Girija, M. and Dhanavel, D. (2010).** Effect of gamma rays, EMS, DES and COH on protein and oil content in soybean. *J. Eco. Bio. Tech.*, **2**(4) : 47-50.
- Rachie, K.O. (1975).** The millets: Importance, utilization and overlook. International Crops Research Institute for the Semi-Arid Tropics, Hyderabad, India, 63 pp.
- Sharma, S.M. (1993).** Utilization of national collection of sesame in India. ICAR-IBPGR Reg. Workshop on sesame evaluation and improve, 28-30th, Sep., Nagpur, India.
- Singh, S., Richharia, A.K. and Joshi, A.K. (1998).** An assessment of gamma ray induced mutations in rice (*Oryza sativa* L.). *Indian J. Genet.*, **58**(4) : 455-463.
- Solanki, I.S. and Sharma, B. (1994).** Mutagenic effectiveness and efficiency of gamma rays, ethyleneimine and N-nitroso-N-ethyl urea in macrosperma lentil (*Lens culinaris* Medik.). *Indian J. Genet.*, **54**(1) : 72-76.
- Subramanian, A., Nirmalakumari, A. and Veerabhadhiran, P. (2011).** Mutagenic efficiency and effectiveness in kodo millet (*Paspalum scrobiculatum* L.) *Madras Agric. J.*, **98**(1-3) : 22-25.
- Swaminathan, M.S. (1969).** Role of mutation breeding in a changing agriculture. In : *Induced mutations in plants*. IAEA, Vienna, pp. 719-734.
- Talebi, A.B. and Talebi, A.B. (2012).** Radiosensitivity study for identifying the lethal dose in MR219 (*Oryza sativa* L. sp. *Indica* Cv. MR219). *Internat. J. Agril. Sci, Res. & Tech.*, **2**(2) : 64-67.
- Thilagavathi, C. and Mullainathan, L. (2009).** Isolation of macro mutants and mutagenic effectiveness, efficiency in black gram [*Vigna mungo* (L.) Hepper]. *Glo. J. Mol. Sci.*, **4**(2) : 76-79.



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