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RESEARCH ARTICLE

Study on induced mutations in m_1 generation in sorghum [*Sorghum bicolor* (L.) Moench]

■ **S.M. Surashe, H.V. Kalpande and S.B. Borgaonkar**

SUMMARY

An investigation was carried out to create the variability generated through induced mutation in two sorghum populations *viz.*, 296 B (*Kharif*) and Parbhani Moti (*Rabi*). Two mutagens *viz*., gamma irradiation (10 kR, 20kR, 30kR and 40kR) and EMS (0.1%EMS, 0.2%EMS, 0.3%EMS and 0.3%EMS) and their combination were used M_1 generation. Mutagenic sensitivity in M_1 generation on the basis of reduced germination and plant survival revealed a dose dependent reaction and differential response of the populations. LD_{50} was found to be 20-30 kR in case of gamma irradiation and 0.3-0.4 per cent in EMS irrespective of the genotype. The irradiated population produced more number of superior segregants in respects of seed yield and its contributing traits compared to other populations. Three dwarf mutant, one brown midrib and tree drought tolerance confirmed from Parbhani Moti.

Key Words : Mutation, Segregants, EMS, LD_{50}, Genetic variability

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Sorghum (*Sorghum* spp.) is cultivated predominantly in USA, China, India and Africa for both human and livestock consumption. In India, Sorghum is cultivated over of 4.10 million ha with an annual production of 4.17 million tonnes of grain with a productivity of 1018 kg/ha (Ministry of Agriculture Government of India, 2018). In India, the productivity of *Rabi* sorghum is very low and highly variable from year to year mainly due to post flowering drought but *Rabi* sorghum is highly valued because of its good grain quality. This crop is usually affected by water stress at both pre and post-flowering stages of development and has the most adverse effect on yield (Kebede *et al*., 2001). Drought tolerance is defined as the relative yield of a genotype compared with other genotypes subjected to same drought stress (Hall, 1993). Drought tolerance depends on the plant developmental stage at the onset of the stress condition, which may happen in sorghum

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during the early vegetative seedling stage, during panicle development and in post-flowering, in the period between grain filling and physiological maturity. Genetic variability for economic traits is the pre-requisite for any successful breeding programmers as the degree of response to selection depends on the quantum of variability. Mutation breeding is recognized as one of the driving force of evolution. Mutation breeding is relatively quicker method for improvement of various crop species. It is an important tool to create variability for quantitatively inherited traits in different plants and is considered as an alternative method to increase genetic variability in plant breeding programmes. It is often used to correct defects in a cultivar which has a set of good agronomic characteristics. Among various physical mutagens such as x-rays, fast neutrons, thermal neutrons, ultraviolet and beta radiation, gamma rays in particular are well known with their effect on the plant growth and development by inducing cytological, physiological and morphological changes in cell and tissues (Thapa, 2004). Gamma radiation is an important tool for inducing the genetic variability, enhancing yield and yield contributing traits. However, the changes brought about by mutations were not very successful in simultaneous improvement of all yield-contributing characters. It is suggested that the application of mutagenic treatments to hybrids may be one of the mean of adding the variability inherent in the cross to that induced by mutation. Complementing the conventional methods, mutation breeding can play a unique role in crop improvement which provides a novel approach to plant breeder for improving the productivity of crop plants. Proves to be the method of choice for obtaining quicker results, when it is desired to rectify small defects in any crop variety. The present investigation was taken up to study the relative effects of physical and chemical mutagens in M_1 generation

MATERIAL AND METHODS

The material for the present study was undertaken at Sorghum Research Station, Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani. The pure seed of two commercial varieties of sorghum [*Sorghum bicolar* (L). Moench] *viz*., 296 B and Parbhani Moti were selected for mutagenic treatment. Selfed seeds (1200) with about 10 ± 1 per cent moisture for each of the two varieties *viz*., 296 B and Parbhani Moti were exposed to 10, 20, 30 and 40 kR dose of gamma rays (CO^{60}) with a dose rate of 2.39 kR per minute at Nuclear Agriculture and Biotechnology Division, B.A.R.C. Trombay, Mumbai, and the same number of untreated seeds of each variety served as control. Seeds treated with chemical mutagens were thoroughly washed in tap water remove the traces of mutagens and were immediately sown in the field of Sorghum Research Station, Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani on dated 12st January 2016 (Summer, 2016) By dibbling one seed $hill^{-1}$ at a distance of 45 cm between rows and 15 cm between seeds. The experiment was laid in a Randomized Block Design (RBD) with three replication. Twelve hundred seeds of each treatment including control (untreated seeds) were sown in a plot size of 6.75 m x 4 m (15 rows of 4 m length) in each replication. Recommended package of practices was followed to raise a good crop. The mean values recorded for various traits in M_1 generation were used for further statistical analysis. The statistical analysis was carried out as per standard method of Analysis of variance for Randomized Block Design (Panse and Sukhatme, 1954).

RESULTS AND DISCUSSION

The results obtained from the present investigation as well as relevant discussion have been summarized under following heads :

Studies in M_1 generation:

Mutagenic sensitivity in generation:

Germination:

The effect of gamma rays, ethyl methane sulphonate (EMS) and their combination on germination in two varieties of sorghum was studied during M_1 generations and results are presented in Table 1. The data revealed that the germination per cent of 296 B and Parbhani Moti was reduced in all the mutagenic treatment as compared to control. The maximum reduction in the germination per cent was observed in gamma rays followed by EMS dose. Gamma rays dose of 40 kR recorded the lowest germination per cent (39.33) followed by 0.4 per cent ethyl methane sulphonate (43), 30 kR gamma rays (50.33), 0.3 per cent EMS (55.33) and their combination 20 kR+ 0.2 per cent EMS (56.66). The highest germination per cent was observed in 0.1 per cent EMS (72.66) followed by 10 kR gamma rays (69.66%). The effect of different mutagenic treatments on germination per cent in variety Parbhani Moti was studied and it was observed that, the lowest germination per cent of 40.66 per cent was recorded in 40 Kr gamma

rays followed by 0.4 per cent EMS (45.33%). Among the all two population the highest germination per cent (73.66%) was observed in 0.1 per cent EMS concentration and lowest (39.33%) in 40 kR gamma irradiation in case of Parbhani Moti genotype.

Plant survival :

The data with respect to plant survival is presented in Table 1. Both gamma irradiation and EMS treatments were effective in reducing the survival in all the two populations. The data revealed that, survival of plants decreased with an increase in dose or concentrations of gamma rays, EMS and their combination in M_1 generation. The maximum survival in M_1 generation was obtained in 0.1 per cent EMS (67%) in 296 B population and 68.33 per cent in Parbhani Moti of 0.1 per cent EMS. The minimum survival rate of 37.66 per cent was obtained in 40 kR dose of gamma rays for 296 B and 38.34 per cent in Parbhani Moti populations.

Estimation of LD_{50} based on germination:

The genotypic differences for both the mutagenic treatments were observed. The genotype 296 B was more sensitive to both the mutagens compared to other population. LD_{50} for seed germination in case of gamma rays was found to be 26.90 kR (296 B) and 28.16 kR (Parbhani Moti) whereas in case of EMS treatments, it was 0.32 per cent (296 B) and 0.33 per cent (Parbhani Moti). This indicated that the LD_{50} dose between 25-35 kR in case of gamma rays and 0.3-0.4 per cent in case of EMS were ideal for mutagenic treatment in case of sorghum.

Induced polygenic variance in M_1 generation:

The induced polygenic variability calculated for four quantitative characters in M_1 generation of two varieties of sorghum cultivars *viz*., 296 B and Parbhani Moti. In general it was observed that the range, variance and co-efficient of variation were increased due to mutagenic treatment over control in all the populations for all characters studied with few exceptions. It cultivar also observed that the M_1 (296 B) progeny variance values for range, variance and co-efficient of variation found significant than M_1 progeny values for all characters.

Earhead length (cm):

The effect of different mutagenic treatments on population variance for earhead length in two cultivars of Sorghum *viz*., 296 B and Parbhani Moti was studied during M_1 generation and results are presented in Table 3. In cultivar 296 B, 0.3 per cent EMS treatment recorded higher variance (16.52), whereas 40 kR dose of physical mutagen induced higher variance (6.91) in Parbhani Moti. Among the combination of mutagenic treatments, 10kR+0.2% EMS induced highest variance (12.31) in 296 B populations. While 20kR+0.2% EMS found effective for highest variance (7.14) in Parbhani Moti population as compared to their control. The co-efficient of variation in cultivar Parbhani Moti, 30 kR gamma rays dose induced highest co-efficient of variation in M_1 (19.48) generation. Whereas among chemical mutagenic treatments the cultivar 296 B, 0.4 per cent EMS

Table 1: Effect of different mutagens on germination and survival populations of M_1 generation in sorghum

Sr. No.	Treatments	296 B		Parbhani Moti	
		Germination %	Survival %	Germination %	Survival %
1.	10 kR	69.66	65	70	67
2.	20 kR	63.33	60.33	64	61.33
3.	30 kR	50.33	50.00	52.66	51.35
4.	40 kR	39.33	37.66	40.66	38.34
5.	0.1 %EMS	72.66	67	73.66	68.33
6.	0.2 %EMS	64.66	62.66	66.33	64.66
7.	0.3 %EMS	55.33	52.30	56.33	55.33
8.	0.4 %EMS	43	40	45.33	44.66
9.	10kR+0.1%EMS	66	62.33	65	62.33
10.	10kR+0.2%EMS	61.66	58.33	63	60.33
11.	20kR+0.1%EMS	58	56	59	57.66
12.	20kR+0.2%EMS	56.66	51.33	53.66	53.33
13.	Wet control	80.33	79	81	80
14.	Dry control	79	78	80.66	79.33

mutagenic treatment recorded maximum co-efficient of variation (17.15) in M_1 generation as compared to control. While combination treatments 20 kR+0.2% EMS found effective treatment (18.54) as against control.

Grain yield per plant (g):

The effect of different mutagenic treatments on population variance for grain yield per plant in two cultivars of sorghum *viz*., 296 B and Parbhani Moti was studied during M_1 generation and results are presented in Table 3. In cultivar 296 B, 0.3 per cent EMS treatment was recorded highest variance (90.20%) followed by 10 kR gamma rays (86.93%) as against control. Among the combination mutagenic treatments, 20kR+0.2%EMS induced highest variance (72.79%) in 296 B populations as compared to control. The treatment 10kR gamma rays induced highest co-efficient variation (30.38) in 296 B of M_1 generation. Whereas among chemical mutagenic treatments 0.4 per cent EMS recorded maximum coefficient of variation (27.48) in M_1 generation as

Table 2 : LD_{50} for germination of sorghum populations

Sr. No.	Populations	Gamma rays (kR)	EMS (%)
		Germination	Germination
1.	296 B	26.90	0.32
2.	Parbhani Moti	28.16	0.33

Table 3 : Range, mean, variance and co-efficient of variance (CV) for Earhead length and grain yield per plant in M_1 generation of sorghum

Sr. No.	Treatments	Earhead length				Grain yield per plant			
		Range (cm)	Mean ± SE	Variance	CV (%)	Range (cm)	Mean ± SE	Variance	CV (%)
296-B									
1.	10 kR	16-26	22.03+0.57	9.90	14.28	17-47	30.70+1.70	86.93	30.38
2.	20 kR	15-27	23.53+0.52	8.26	12.21	17-47	29.03+1.53	70.17	28.85
3.	30 kR	14-28	22.33+0.62	11.54	15.21	23-48	37.57+1.27	48.53	18.54
4.	40 kR	15-28	24.07+0.59	10.41	13.41	14-47	32.67+1.63	79.75	27.34
5.	0.1 % EMS	14-28	23.20+0.67	13.34	15.74	20-46	33.10+1.46	64.02	24.17
6.	0.2 % EMS	14-28	22.03+0.59	3.34	14.57	20-45	33+1.49	66.28	24.67
7.	0.3 % EMS	15-27	21.03+0.74	16.52	19.32	18-48	37.07+1.73	90.20	25.62
8.	0.4 % EMS	16-28	22.63+0.62	11.46	14.97	17-50	33.07+1.66	82.55	27.48
9.	10 kR + 0.1 % EMS	16-26	23.03+0.51	7.76	12.09	19-47	32.33+1.40	59.20	23.80
10.	10 kR + 0.2 % EMS	14-29	22.03+0.64	12.31	15.92	20-48	29.33+1.37	56.71	25.67
11.	20 kR + 0.1 % EMS	15-28	24.26+0.56	9.37	12.62	18-48	31.73+1.36	55.79	23.54
12.	20 kR + 0.2 % EMS	14-28	22.12+0.62	11.62	15.41	19-48	37.37+1.56	72.79	22.83
13.	Wet control	18-25	23.67+0.58	10.18	13.37	22-42	30.30+1	29.73	17.66
14.	Dry control	21-25	23.20+0.38	4.23	11.50	25-41	35.53+0.84	20.95	12.88
Parbhani Moti									
1.	10 kR	11.00 -16	13.23±0.33	3.36	13.85	24.00-50	37.60±1.25	46.80	18.19
2.	20 kR	11.00-18	14.10±0.43	5.62	16.80	20.00-54	49.00±1.71	88.14	19.16
3.	30 kR	10.00-17	13.03±0.46	6.45	19.48	24.00-53	42.00±1.25	46.76	16.28
4.	40 kR	11.00-19	13.70±0.48	6.91	19.18	28.00-54	40.37±1.19	42.65	16.18
5.	0.1 % EMS	11.00-17	14.03±0.32	3.14	12.62	35.00-55	44.27±1.07	34.62	13.29
6.	0.2 % EMS	11.00-18	13.40±0.34	3.49	13.94	28.00-55	46.73±1.36	55.79	15.98
7.	0.3 % EMS	12.00-19	15.83±0.44	5.71	15.10	25.00-58	45.00±1.35	54.62	16.42
8.	0.4 % EMS	11.00-19	15.00±0.47	6.62	17.15	22.00-59	40.97±1.62	78.38	21.61
9.	10 kR + 0.1 % EMS	11.00-17	14.00±0.31	2.81	11.97	34.00-55	42.00±1.42	60.14	18.46
10.	10 kR + 0.2 % EMS	11.00-17	14.14±0.35	3.63	13.47	32.00-58	44.00±1.37	56.14	17.03
11.	20 kR + 0.1 % EMS	11.00-17	13.70±0.43	5.67	17.37	25.00-60	48.00±1.68	84.69	19.17
12.	20 kR + 0.2 % EMS	10.00-18	15.01±0.51	7.74	18.54	24.00-58	42.00±1.43	61.03	18.60
13.	Wet control	12.50-16	14.54±0.19	1.04	7.02	38.00-50	44.00±0.82	20.21	10.22
14.	Dry control	12.00-15	13.95±0.13	0.53	9.24	39.00-52	45.70±0.86	22.08	10.28

compared to control. While in case of combination treatment 10kR+0.2% EMS was found effective treatment (25.67) as against control. In case of Parbhani Moti, 20 kR was recorded highest variance (88.14), whereas 20 kR+0.1% EMS treatment induced more variance (84.69) as against control. Among the chemical mutagenic treatments, 0.4 per cent EMS induced highest variance (78.38) as compared to control. The co-efficient of variation in Parbhani Moti, 0.4 per cent EMS treatment induced highest co-efficient variation (21.61) in M_1 generation. Whereas among combination of mutagenic treatments 20kR+0.1%EMS recorded maximum co-efficient variation (19.17) in M_1 generation as compared to control. Among the gamma rays treatments 20kR dose was found effective co-efficient of variance (19.16) as against to control. The different mutagenic treatments of gamma rays, EMS and their combination were effective in affecting the survival of the plants in M_1 generation in both cultivars. However, the reduction in survival was more pronounced in M_1 generation, in 296 B and Parbhani Moti. The minimum survival was recorded in 40 kR gamma rays and 0.4 per cent EMS concentration which means the higher dose or concentrations of mutagen had drastic effect on survival of the plants.

The present findings in respect of dose dependent reduction in survival, mutagen differences and differential response of populations to mutagens are in conformity with the earlier reports of Harris *et al*. (1965); Moradi *et al*. (2009); Suthakar *et al*. (2014) and Vishnu Bhala and Verma (2018) in sorghum. The studies on germination and survival in the treated populations assumes importance due to its utility in elucidation of LD_{50} values (Gustafsson, 1944). In the present investigation, LD_{50} was found to be ideal around 30 kR in case of gamma rays and 0.3 per cent of EMS for mutagenic treatments in case of sorghum irrespective of the genotypes. This is in agreement with the finding of Imam (1979) and Larik *et al*. (2009). The findings of Veenakumari (1994) indicated that sorghum populations tolerate relatively higher doses (above 30-40kR of gamma rays and around 0.4% EMS) as against the indications in the present investigation.

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