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MUTAGENIC EFFECT ON GROWTH PARAMETERS OF FOXTAIL MILLET [SETARIA ITALICA (L.) BEAUV.] IN M_1 GENERATION

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ABSTRACT

Among the various plants breeding technique induced mutation using physical and chemical mutagens play a great achieving role in current days in improving economic value of the crop with genetic modification. In the present investigation foxtail millet [*Setaria italica* (L.) Beauv.] seeds were treated with different doses of gamma rays (GR) ranging from 200, 300 and 400Gy, varying concentration of ethyl methane sulphonate (EMS) 0.2, 0.3 and 0.4% and sodium azide (SA) 0.02, 0.03 and 0.04%. Variations in the germination percent of seeds, seedling height, seedling injury and survival of plant at maturity of foxtail millet were recorded in M_1 generation. The effects of the mutagenic treatments on quantitative traits resulting in reduction in traits such as percent seed germination except 200Gy (86.13%), 0.2% EMS (85.96%) and 0.02% SA (87.21%), seedling height except 200Gy (7.02cm), 0.2%EMS (7.12cm) and 0.02%SA (6.96cm) while survival of plant at maturity except 200Gy (82.94%), 0.2% EMS (81.73%) and 0.02%SA (83.02%) but increases seedling injury except 200Gy (-02.03%), 0.2% EMS (-03.49%) and 0.02%SA (-01.16%) was observed in treated M_1 generated plants.

Key words: Mutagens, EMS, SA, foxtail millet, seedling injury.

INTRODUCTION

Foxtail millet [*Setaria italica* (L.) Beauv.] locally called as *Rale*. It belongs to family Poaceae. It is cultivated in arid and semiarid parts due to its health benefits and balance of nutrients, such as starch, protein, dietary fibers, fat, vitamins, good yield and adaptation to different biotic and abiotic stresses like salinity, drought as well as fungal diseases (Lata et al., 2013). Foxtail millet have many important characteristics, like its short stature, short life cycle, good seed production, self-compatibility, true diploid nature, high photosynthetic efficiency, small genome size, as well as tolerance to biotic & abiotic stress (Sun et al., 2019). Now a days millet grains is receiving interest from food scientists, technologists, and nutritionists due to its valuable contribution to national food security as well as potential health benefits (Saleh et al., 2013).

The foxtail millet is a valuable crop and considered as a good therapeutic diet. There fore this crop need to be cultivated in large scale production having good characters by using improved techniques such as mutation breeding. The millet grains is used as feed for love birds, grained flakes and malt based product (Anittha and Mullainathan, 2018). Whole-grain cereals protect the body against age-related diseases such as diabetes, cardiovascular diseases, and some cancers (Fardet et al., 2008). The germinated seed is astringent, digestive, emollient and stomachic. It is used in the treatments such as poor digestion and food stagnancy in the abdomen, as well as cholera and fever (Duke, 1985). Millet grains are processed and consumed by using processing methods such as decorticating, malting, fermentation, roasting, flaking, and grinding, it improve their edible, nutritional, and sensory properties (FAO 2012). The nutritional composition of millet grain per 100g edible portion is: water- 12g, protein- 11.2g, fat- 4.0g, carbohydrate- 63.2g, crude fibre- 6.7g, Ca- 31mg, Fe- 2.8mg (FAO, 1995).

MATERIALS AND METHODS

The seeds of foxtail millet [*Setaria italica* (L.) Beauv.] were procured from local market of Manchar, Tal. Ambegaon, Dist.- Pune- 410503 (M.S.) India. Gamma rays (GR), ethyl methane sulphonate (EMS) and sodium azide (SA) were used as mutagens in present investigation for seed treatments. Gamma radiation from ^{60}Co source fixed in the gamma cell 200 installed at Department of Chemistry, Savitribai Phule, Pune University, Pune was used in the present work. Dry, healthy and uniform seeds of foxtail millet with moisture content of 10 to 12 % were treated with 200, 300 and 400 Gy. Ethyl methane sulphonate ($CH_3SO_2OC_2H_5$) molecular weight 124.16, manufactured by Sigma chemical Co. Ltd. USA was used for the seed treatment of foxtail millet. Various concentrations of EMS (0.2, 0.3 and 0.4%) were prepared in distilled water. Sodium Azide is a chemical mutagen, used for induction of mutations in the crop plants. Different conc. of SA (0.02, 0.03 and 0.04%) was prepared in distilled water.

The experiments were conducted to determine the lethal dose (LD_{50}), suitable concentrations of EMS, SA and duration of seed treatment. The doses of gamma rays, 200, 300 and 400 Gy, EMS 0.2, 0.3 and 0.4% while SA 0.02, 0.03 and 0.04% were finally selected for the seed treatment and the duration fixed was four hours. Selected seeds were soaked in distilled water for 8 hours and the wet seeds were treated with different concentrations of EMS and SA for four hours. The untreated seeds served as control. For each treatment 180 seeds were used. The treated seeds washed thoroughly with tap water for one hour to leach out the residual chemicals. From each treatment 30 seeds was used for seed germination in laboratory. Three replications with 10 seeds per replication kept in petri dishes, containing seed germination paper, were used for recording seed germination, seedling height on seventh day. The remaining lot of treated seeds (150) from each treatment was used for raising M_1 generation in field. The field experiments were conducted on the experimental plot at Department of Botany. The soil type of the experimental field was slightly deep, fine and with good drainage. The average minimum temperature was recorded as 17.63^0C and maximum 32.73^0C with average annual rainfall 641.03mm. The experiments were carried out following RBD design. Each plot had 50 plants. The distance between two rows and two plants was 45 X 30cm.

Observations on M_1 generation : The number of seeds showing emergence of the radical and plumule was used to calculate percent seed germination. On seventh day of sowing, 5 seedlings from control and each treatment were randomly selected for measuring the root and shoot length and the average values were recorded in table. Reduction in the mean seedling length as compared to the control was regarded as seedling injury and expressed as percentage.
% seedling injury = (Control seedling height - Treatment seedling height) / Control seedling height X 100.
Survival percent was calculated by scoring the number of plants attaining maturity (45days).

STATISTICAL ANALYSIS

The data were summarized as the means of three replicates with standard deviation as the measures of variability. One-way ANOVA test was performed to determine significant differences due to various treatments. Fisher's LSD (Least significant difference) was used as post hoc test to as certain significant differences among treatments at p= 0.05. Statistical analysis and graphical data presentations were carried out by using IBM SPSS (ver.25).

RESULTS AND DISCUSSION

The results on seed germination percent, seedling injury and survival of plant at maturity in M_1 generation of foxtail millet were recorded in Table-1. Seed germination in control and mutagen treatments clearly indicated that it was decreased in all the treatments except in lower treatments such as 200Gy, 0.2%EMS and 0.02%SA as compared to control. The mutagens had exerted adverse effects on seed germination. The percent seed germination decreased from 86.13% to 56.34% in GR except 200Gy (86.13%), 85.96% to 58.07% in EMS except 0.2%EMS (85.96%) and 87.21% to 59.38% except 0.02% (87.21%) in SA. The maximum (50%) decrease in percent seed germination was observed with GR treatment 400Gy (56.34%), EMS 0.4% (58.07%) and in SA 0.04% (59.38%). The results of present study have clearly shown that foxtail millet was sensitive to all the mutagens except 200Gy, 0.2%EMS and 0.02SA. 200Gy, 0.2%EMS and 0.02SA showed increase in seed germination percent over control. Reduction in seed germination with increasing dose/ conc. of mutagens was reported in finger millet (Bolbhat and Thikekar, 2020, black turtle bean (Bolbhat et al., 2020) and in barnyard millet (Bolbhat and Bhalekar, 2020). Results clearly indicates that lower doses/conc. of mutagens like GR, EMS and SA showed increase in seed germination percent.

Data recorded in table 1 indicates that doses / conc. of mutagen treatments showed negative effect on seedling height except in lower treatments. Minimum seedling height (3.63cm) was noted in 0.04%SA, 0.4%EMS (3.86cm) and 400Gy (4.59cm). Treatments such as 200Gy (7.02cm), 0.2%EMS (7.12cm) and 0.02%SA (6.96cm) showed increase in seedling height than control (6.88cm).

Table 1 : Effect of mutagens on growth parameters in M_1 generation of foxtail millet

Treatments	Germination (%)	Root length (cm)	Shoot length (cm)	Seedling height (cm)	Seedling injury (%)	Plant survival (45 days) (%)
Control	85.64±11.99	3.25±0.46	3.63±0.51	6.88±0.96	00.00±0.00	81.25±11.38
200Gy	86.13±6.89	3.32±0.27	3.70±0.30	7.02±0.56	-02.03±0.16	82.94±6.64
300	70.27±7.73	2.53±0.28	2.84±0.31	5.37±0.59	28.95±3.18	65.07±7.16
400	56.34±7.32	1.92±0.25	2.67±0.35	4.59±0.60	33.28±4.33	53.16±6.91
0.2 % EMS	85.96±12.03	3.40±0.48	3.72±0.52	7.12±1.00	-03.49±0.49	81.73±11.44
0.3	72.53±5.08	2.36±0.17	2.57±0.18	4.93±0.35	28.34±1.98	65.47±4.58
0.4	58.07±4.65	1.77±0.14	2.09±0.17	3.86±0.31	43.90±3.51	51.19±4.10
0.02% SA	87.21±13.08	3.27±0.49	3.69±0.55	6.96±1.04	-01.16±0.17	83.02±12.45
0.03	74.46±10.42	2.47±0.35	2.32±0.32	4.79±0.67	30.78±4.31	68.24±9.55
0.04	59.38±5.34	1.68±0.15	1.95±0.18	3.63±0.33	47.24±4.25	51.68±4.65
SEM±	07.33	00.27	00.30	00.57	02.34	06.88
F-value	05.77	12.74	11.28	11.68	14.77	07.50
P-value	00.01	00.01	00.01	00.01	00.01	00.01
$LSD_{0.05}$	15.97	0.59	00.65	01.24	05.10	14.99

Data are means of three replicates ± standard deviation. Significant difference due to treatments was assessed by Fisher's LSD as a post-hoc test.

Seedling injury at M_1 showed that all mutagenic treatments except lower treatments were highly injurious to the seedlings. SA treatments had caused highest seedling injury, followed by EMS and the gamma radiation. The seedling injury increased with the increase in doses/ concentrations of mutagenic treatments. Maximum seedling injury (47.24%) was observed in 0.04%SA, followed by 0.4% (43.90%) in EMS and 400Gy (33.28%) in GR. Lower treatments such as 200Gy (-02.03%), 0.2%EMS (-03.49%) and 0.02%SA (-01.16%) showed negative seedling injury. The seedling injury significantly increased with the increase in doses/ conc. of mutagenic treatments except lower treatments in foxtail millet. Similar results has been reported by earlier researchers Uttarde et al., (2020) in sesame, Bolbhat and Bhalekar, (2020) in barnyard millet, Senapati *et al*., (2008) in blackgram.

The results on the effects of mutagens showed that in all the mutagenic treatments, survival % was decreased but increased in lower treatments over the control. There was decrease in the survival % with increasing dose/ conc. of gamma radiation, EMS and SA except 200Gy (82.94%), 0.2%EMS (81.73%) and 0.04%SA (83.02%). The lowest survival % at the higher treatments was noted (51.19%) in 0.4%EMS, (51.68%) in 0.04%SA, and (53.16%) in 400Gy as compared to control (81.25%). Results indicates that lower dose/ conc. of mutagens showed increasing survival percentage while all other showed reduced the rate of survival at maturity. Awate and Bolbhat (2020) in horsegram, Bolbhat and Bhalekar, (2020), Ramesh et al., (2019) in barnyard millet, Bolbhat and Thikekar, (2020) in finger millet supported the above findings.

CONCLUSION

Seed germination percent and seedling height was inhibited due to increasing doses/ concentrations of mutagens except lower mutagenic treatments. All three mutagens (GR, EMS and SA) were effective in inducing seedling injury in M_1 generation. The rate of seedling injury percent increased but lower treatments showed decrease in seedling injury over control. The rate of survival of plants at maturity was highly reduced with increasing dose/conc. of mutagens.

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