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Identification of Mutants in Groundnut *(Arachis hypogaea* L.) induced through Sodium Azide

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ABSTRACT

The present investigation was carried out to determine the mutagenic effect of Sodium Azide on morphological traits of Kadiri-6, Gujarat Gold, Haritha Andhra and Kadiri Amaravati (*Arachis hypogaea L.)* in M_2 generation. Pre-soaked seeds were treated with different concentrations w/v (0.1, 0.15, 0.20, 0.40) of sodium azide for 17 hrs at room temperature. The experiment was carried out with four doses of mutagen (sodium azide) for four different varieties along with their respective controls in the Randomized block design with three replications. The observation of growth parameters of both M_1 & M_2 mutants of Kadiri-6, Gujarat gold, Haritha Andhra and Kadiri Amaravati showed a significant variation in different traits such as germination percentage, survival percentage, seedling height, days to maturity, days to flowering, number of sterile plants, plant height at maturity, number of pods per plant, sound mature kernels and kernel yield per plant. Most of the traits decreased with an increase in the concentration dosage of sodium azide. The result of the study shows that sodium azide majorly affected the above-mentioned traits, it has significant influence on all the traits under study.

Key words : *Groundnut, Mutant, Sodium Azide, Kadiri-6, Gujarat Gold, Haritha Andhra, and Kadiri Amaravati.*

Introduction

Groundnut (*Arachis hypogaea L.*) is a member of the Papilionaceous, subfamily of the Fabaceae family. Groundnut is native to Brazil in South America and is presently cultivated throughout tropical, subtropical, and warm temperate regions of the world. It is a self-pollinated crop and allotetraploid with 2n=4x=40 chromosomes. (Kochert *et al*., 1996). Globally groundnut covers 295 lakh hectares with the production of 487 lakh tonnes with a productivity of 1647kg per hectare (FAOSTAT, 2019). India ranks first in groundnut crop acreage and second in production of groundnut in the world with 101 lakh tonnes with a productivity of 1816 kg/ha in 2020-21(agricoop.nic.in). Groundnut seeds harbor high-quality edible oil (50%), easily digestible protein (25%), and carbohydrates (20%). Besides it is an important source of minerals like phosphorus, calcium, zinc, and vitamins *i.e.,* A, C, E, K, thiamine and niacin. Groundnut is richer in protein than meat, egg and other vegetables. Freshly roasted groundnut with jaggery and goat milk is very nutritious for growing children, pregnant women and nourishing mothers. It builds resistance against hepatitis and tuberculosis.

A mutation is a sudden heritable change in the genetic material of an organism. Several physical and chemical mutagens have been used for the induction of useful mutations in several crops. The approach of mutation breeding is to generate new genetic variability in a given crop species when

there is a little variation for a certain trait (Yadav *et al.*, 2007).

The induced mutation may be an alternative, complementary or unique technique in creating genetic variability as in cross-breeding. The range of variability released through the mutation technique depends on the genotype or kind and dose of mutagen employed (Sarma, 1975; Reddy *et al*., 1977; Levy *et al*., 1979).

Sodium azide (SA) is well known heavy metal enzyme with influences on metabolism and respiration of the living cell. SA was found to decrease the cellular level of calmodulin, which is a calcium-binding protein participating in signal transduction and cell division. Different mutation breeding studies on groundnut involve seed treatment of a popular germplasm material to generate mutant genotype with additional desirable characteristics with different mutagens at various levels of concentrations.

Materials and Methods

The present experiment was carried out during the *Kharif* seasons of 2020 and 2021 *i.e.*, M_1 &M_2. It was conducted at the Crop Research Centre (CRC), Deptt of Genetics and Plant Breeding, School of Agriculture, ITM University, Gwalior, MP, India. It is located at a latitude of 26°14′ N and a longitude of 78° 14′ E with an elevation of 206 m above the mean sea level. The germplasm of four Spanish bunchy type varieties of groundnut *i.e*., Kadiri-6, Gujarat Gold, Haritha Andhra, and Kadiri Amaravati were collected from the Agriculture Research Station, Kadiri, Andhra Pradesh. The seeds were treated with the different concentration levels of mutagen *viz*. Sodium Azide (NaN_3). The four concentration (0-control) 0.1, 0.15, 0.20, 0.40 w/v. The different concentrations of mutagens were prepared by dissolving 0.1, 0.15, 0.20, and 0.40 g of sodium azide in 100 ml of distilled water respectively. Before treatment seeds were pre-soaked in distilled water for two hours as it allows the cells to reach an active metabolic state when they are relatively more sensitive to mutagenic action and then the seeds of four varieties were treated with different concentrations (0.10, 0.15, 0.20 and 0.40) of SA for 17hrs at room temperature (Joshi, 2017). After treatment, the seeds were washed thoroughly with distilled water. The treated seeds of groundnut were sown in Randomized Block Design (RBD) with three replications. The sowing was done by dibbling seeds in rows with a spacing of 60x10 cm and recommended package of practices were adopted for optimum crop growth. The traits under study included germination percentage, survival percentage, seedling height, days to maturity, days to flowering, number of sterile plants, plant height at maturity, number of pods per plant, sound mature kernels, and kernel yield per plant.

Data collected during crop growth for the traits under study was subjected to analysis using R Studio (Software). Plants that were expected to be mutants based on morphological appearance were selected and grown in M_2 generation. For each putative mutant from M_1, the plants were cultivated in three replications in the M_2 generation. The M_2 generation was planted with a 60 x 10 cm plant to plant and row to row distance. To accurately record the data, a single plant per hill was raised. All of the agricultural procedures were kept up to date to raise a decent crop.

Results and Discussion

The effect of the different concentrations of sodium azide on different traits includes germination percentage, survival percentage, seedling height, days to maturity, days to flowering, number of sterile plants, plant height at maturity, number of pods per plant, sound mature kernels, and kernel yield per plant. The frequency of mutants on different traits is presents in Table 1.

M_1 Generation

In M_1 generation the result showed that different treatments of sodium azide such as T_1-T_4 (0.1, 0.15, 0.2 & 0.4%) significantly influence different traits of the groundnut when compared to the control which is untreated in the present investigation this shows that magnitude of the parameters such as germination percentage, survival percent, seedling height, days to maturity, days to flowering, number of sterile plants, plant height at maturity, number of pods per plant, sound mature kernels and kernel yield per plant, decreased with the subsequent increase in the dose of mutagen (Sodium Azide) which was depicted in Table 1.

M_2 Generation

The mean performance of the following traits has the effect of mutagenic treatment as compared to

control is mentioned below.

Germination percentage

In contrast to the control (93.67 percent), of variety kadiri-6, it was discovered that a sodium azide dose at a concentration of 0.15 percent was the most effective. The germination was significantly reduced in the M_1 and M_2 generations, by 89 and 85%, respectively. This outcome is in sync with Khadre *et al.*, (2015).

No. of pods per plant

In the Kadiri-6 and Kadiri Amaravati varieties, the number of pods per plant decreased as the dose's concentration increased. The 0.20 percent dose in Kadiri -6 and the 0.40 percent dose in Kadiri Amravati demonstrates greater efficacy. In Kadiri-6, the M_1 and M_2 generations saw reductions in the number of pods produced per plant of 25 and 23.33 compared to control (28) and 25 and 25 in Kadiri Amaravati, with respect to the control (32.33). The outcome was comparable to that of Ahloowalia *et al.*, (2004) and Arulbalachandran (2006).

Kernel yield per plant

The Kernel yield per plant declined with increasing dosage concentration in the Kadiri-6, Gujarat Gold, and Kadiri Haritha Andhra cultivars. More effective doses include 0.15 percent in Kadiri-6, 0.20 percent in Gujarat Gold, and 0.10 percent in Kadiri Haritha Andhra. The kernel yield per plant decreased by 10.30 and 7.67 percent in Kadiri-6 for the M_1 and M_2 generations compared to the control (10.97), 8.47 and 6.80 percent in Gujarat Gold compared to the control (9.20), and 9.63 and 6.63 percent in Kadiri Haritha Andhra compared to its control (10.10), respectively. Similar results were obtained by Amarnath *et al.*, 1991 and Balakrishnan, (1991).

Sound mature kernels

It was found that a sodium azide dose at a concentration of 0.15 percent for Gujarat Gold and 0.20 percent for Kadiri Amaravati were the most effective, as opposed to the control (81.67 & 77.00). The Sound mature kernel was greatly lowered in the M_1 and M_2 generations, by 75.67 and 74.60 in Gujarat gold and 74.00 and 71.67 in Kadiri Amaravati, respectively.

Table 1. Analysis of variance for M_1

Treatment	Germination (%)	Survival Percentage	Seedling height	Days to flowering	Days to maturity	Sterile plants	Plant height at Maturity	No.of. Pods per plant	Sound mature kernel	Kernel yield per plant
Kadiri-6 Control	93.67	91.00	16.37	28.00	107.33	0.03	31.50	28.00	74.33	10.97
SA-0.10	88.33	80.33	16.63	27.67	110.00	3.33	32.03	29.33	72.33	10.60
SA-0.15	89.00	82.67	14.10	35.00	104.67	3.13	29.57	28.00	72.00	10.30
SA-0.20	78.67	74.33	13.97	37.00	118.67	3.93	25.40	25.00	68.67	9.90
SA-0.40	76.33	65.00	11.00	39.67	125.67	5.57	23.33	22.67	61.00	6.03
G.Gold Control	93.67	93.33	16.90	30.00	109.67	0.00	34.90	28.67	81.67	9.20
SA-0.10	91.33	84.00	15.50	29.00	108.00	3.13	33.77	30.00	77.67	8.83
SA-0.15	84.00	82.67	14.70	33.00	108.00	2.80	38.57	25.33	75.67	8.40
SA-0.20	74.00	75.67	13.80	35.67	116.33	4.93	33.27	22.00	70.67	8.47
SA-0.40	74.00	66.33	11.47	37.00	122.33	6.37	27.17	22.33	69.67	5.83
H.AndhraControl	92.33	91.67	16.60	29.00	111.33	0.00	32.67	30.67	77.33	10.10
SA-0.10	87.00	89.00	17.03	25.00	113.00	3.37	33.23	27.33	77.67	9.63
SA-0.15	88.67	78.67	16.53	31.00	109.33	3.37	27.50	28.33	71.33	5.97
SA-0.20	88.00	72.67	13.37	33.33	118.33	4.13	29.00	22.33	70.00	4.63
SA-0.40	65.67	66.00	11.70	36.67	121.33	7.90	25.33	19.33	66.67	5.00
Kadiri Amaravati Control	92.33	92.33	16.73	31.33	106.67	0.00	33.90	32.33	77.00	10.73
SA-0.10	82.33	80.67	17.40	30.00	107.33	3.07	25.63	29.00	72.33	6.13
SA-0.15	90.00	80.67	13.77	27.33	106.00	4.47	33.40	24.33	79.00	5.40
SA-0.20	77.33	68.33	13.43	33.67	116.33	6.47	25.43	23.67	74.00	6.60
SA-0.40	73.67	63.33	11.07	34.67	121.67	6.17	29.03	25.00	70.00	5.33
SEM	2.22	2.18	0.80	1.56	2.18	0.37	1.94	1.70	1.77	0.55
CD	6.35	6.23	2.28	4.47	6.25	1.06	5.56	4.87	5.07	1.59
CV (%)	4.57	4.78	9.43	8.40	3.35	17.73	11.13	11.26	4.21	12.16

Table 2. Comparison of Control, M_1 and M_2

Character	Mutagen	Variety	Dose	Control	M_1	M_2
Germination (%)	S.A	Kadiri-6	0.15%	93.67	89.00	85.00
No. of pods per plant	S.A	Kadiri-6	0.20%	28	25.00	23.33
Kernel yield per plant	S.A	Kadiri-6	0.15%	10.97	10.30	7.67
Plant height at maturity	S.A	G. Gold	0.10%	34.90	33.77	28.77
Sound mature kernel	S. A	G. Gold	0.15%	81.67	75.67	74.67
Kernel yield per plant	S. A	G. Gold	0.20%	9.20	8.47	6.80
Days to maturity	S. A	H. Andhra	0.20%	111.33	118.33	120.67
Kernel yield per plant	S. A	H. Andhra	0.10%	10.10	9.63	6.63
Days to maturity	S. A	K.Amaravati	0.10%	106.67	107.33	117.67
No. of pods per plant	S. A	K.Amaravati	0.40%	32.33	25.00	25.00
Sound mature kernel	S. A	K.Amaravati	0.20%	77.00	74.00	71.67

This result is consistent with Arulbalachandran (2006).

Other traits

The traits include Days to maturity and plant height at maturity shows the variation due to mutagenic effect which was shown in Fig. 1.

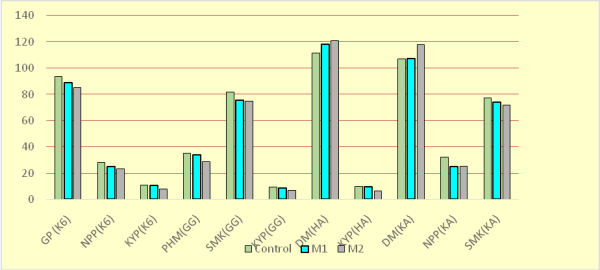

Fig. 1. Comparison of Control with M_1 and M_2

Days to maturity was increased in M_1 and M_2 generations with the increase in dose of mutagen to 118.33 and 120.67 in varieties of Kadiri Haritha Andhra and 107.33 and 117.67 in Kadiri Amaravati. It was found that a sodium azide dose at a concentration of 0.20 percent for Kadiri Haritha Andhra and 0.10 percent for Kadiri Amaravati were the most effective, as opposed to the control (111.33 & 106.67).

The plant height at maturity was decreased in the variety Gujarat gold at the dose of mutagen 0.10 percent in M_1 and M_2 generation by 33.77 and 28.77 as compared to Control (34.90). Similar conclusions were reached by H. Zhang, H. Miao, and M. Ju (2019) as well.

Conclusion

Mutations can be used as a tool for gene functional studies and to create genetic variability (Micke,1979; Rangrao, 2013).

In the M_1 generation, the result shows that the traits such as germination percentage, survival percentage, seedling height, days to maturity, days to flowering, sterile plants, plant height at maturity, number of pods per plant, sound mature kernels, and Kernel yield per plant, are affected with the increase in the dose of mutagenic treatment. In M_2 generations it shown that sodium azide significant effect on germination percentage, no of pods per plant, kernel yield per plant, plant height at maturity, sound mature kernel, and days to maturity along with M_1 generation.

Therefore, it shows mutagenic effect in groundnut which is helpful for further study.

Conflict of Interest

No case of conflict will be established in case any of the author cites the materials present in this research paper. All the authors have contributed in the formation of this research manuscript.

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