The Pharma Innovation Journal 2023; 12(12): 3743-3746

www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2023; 12(12): 3743-3746

www.thepharmajournal.com
Received: 08-09-2023
Accepted: 16-10-2023

Mutagenic effectiveness and efficiency of Sodium Azide in groundnut (*Arachis hypogaea* L.) in M_2 generation

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Abstract
The present investigation was carried out with an objective of assessing the response of two selected groundnut genotypes *viz*., ICGS-76 and Rajashree to different doses of sodium Azide (0.2%, 0.4% and 0.6%). The relative effectiveness and efficiency was assessed from the data on biological damage and frequency of chlorophyll and visible mutants in M2 generation. The mutagenic effectiveness and efficiency were found to be higher for lower and intermediate doses of mutagen in both varieties. Among the two varieties ICGS-76 was found to be more sensitive to mutagenic treatment than Rajashree variety.

Keywords: Effectiveness, efficiency, groundnut, sodium Azide

Introduction
The cultivated groundnut (*Arachis hypogaea* L.) is an annual herb indigenous to South America. Groundnut is a self-pollinating plant which produces flowers above ground. Following fertilization, the pegs go toward the soil, where they develop into seed pods. It is an important oilseed legume crop adapted to a diversity of soils and temperature and is grown around the world. In India, the crop is farmed on around 50 lakh hectares, yielding 80–85 lakh MT. In India, about 80% is used for oil extraction, 11% as seeds, 8% as direct food and 1% for export to other countries (Sharma *et al*., 2021) [15]. The groundnut seeds can be eaten raw as well as cooked. They taste nutty and have a sweet, crispy texture. Because groundnut is a self-pollinating crop with little genetic variation, genetic advancement is crucial for the expansion of the groundnut industry. Crop improvement by mutagenesis has been applied in a number of crops for incorporation of desirable traits in existing plant varieties or for creation of new cultivars. The primary goal of mutation breeding is increasing genetic variation. A number of physical and chemical mutagens have been used for inducing beneficial mutations in a number of crops. Sodium azide is a well-known respiration inhibitor. It prevents the activity of the enzymes catalase and peroxidase (Nilan *et al*., 1973) [11]. This chemical was found to be a potent mutagen in inducing mutants in crop plants (Niknejad *et al*., 1972 and Nilan *et al*., 1973) [9, 11]. The effectiveness, efficiency, plant material, mutagen dose, and duration effects the outcome of mutation breeding (Goyal *et al*., 2019) [6]. The optimum dose of mutagen can be revealed by determining its mutagenic effectiveness and efficiency.

Materials and Methods
During the *Kharif* season of 2021 two varieties of groundnut ICGS-76 and Rajashree were treated with individual doses of 0.2% NaN_3, 0.4% NaN_3 and 0.6% NaN_3. The treated seeds were sown as M_1 generation. The Seeds were harvested from all the treatments of M_1 generation including controls of both varieties. The M_2 generation was raised from the bulk seeds of selected M_1 plants and was laid out in FRBD in three replications along with control at the Experimental Field of the Department of Genetics & Plant Breeding, College of Agriculture, Central Agricultural University, Imphal during *kharif* season of 2022. All the recommended package of practices was followed as and when necessary to raise a good crop of groundnut during the period of investigation. The biological damage (injury and lethality) was computed as the percentage reduction in seedling height and survival respectively. The respective control and treatment progenies were screened several times for chlorophyll and morphological mutations throughout the crop season. Mutation frequency was calculated as percentage of mutated M_2 progenies for both chlorophyll and morphological mutations in each treatment.

Mutagenic effectiveness and efficiency were calculated on the basis of formula suggested by Konzak *et al*., (1965) [8].

Biological damage: For measuring the biological damage, two different criteria were used injury and lethality. Injury is estimated as the ratio of rate of mutation to percentage of seedling height reduction while lethality is estimated as the ratio of rate of mutation to percentage of survival reduction.

Results and Discussion

Mutation frequency

A reliable measure of the genetic impact of mutagens is the frequency of chlorophyll and viable mutants in the M_2 generation (Nilan *et al.*, 1961) [10].

Two types of chlorophyll mutation variegated and viridis along with various viable mutants such as dwarf fertile, dwarf sterile and spreading type mutants were identified. The highest identifiable mutation frequency was recorded from 0.6% NaN_3. Among the cultivars/varieties studied, the highest mutation frequency was recorded from ICGS-76. The present result suggested that the groundnut cultivars/varieties responded differentially to different doses of NaN_3 for the production of mutations.

Chlorophyll mutants

Two kind of chlorophyll mutants were identified in accordance with the classification of Gustafsson (1940) [7] and Blixit (1961) [1].

Viridis

These are viable mutants characterized by light green leaves, plants were slow growing and had a low seed yield.

Variegated

These plants were identified by their leaves having patches of yellow, yellowish white and light green colour.

Viable mutants

Viable mutations were categorized by Gaul (1964) [5] as macro and micro mutations, and by Swaminathan (1964) [17] as macro mutations and systematic mutations. There can be a significant phenotypic shift connected with the mutational event. In the current investigation, some morphological (viable) mutants *viz*. dwarf fertile, dwarf sterile and spreading type were observed in M_2 generation with different doses of sodium azide.

Dwarf fertile

Dwarf fertile mutants were isolated from both ICGS-76 and Rajashree varieties. Dwarf plant with compact nature are desirable as they avoid wastage of plant's energy in excessive vegetative growth, because of which more of the dry matter can be partitioned toward pod development (Swaminathan and Jain, 1973, Duncan *et al*., 1978) [18, 2].

Dwarf sterile

Reduced plant height, compact plant stature, and sterility were the defining traits of these mutants. These mutants were observed in ICGS-76 variety. Pollen development is controlled by a sequential expression of gene specially expressed in reproductive tissues and is highly susceptible to the cellular environment. Any imbalance of the expression of the genes related to anther and microspore development will lead to abnormalities and disable male gametogenesis (Nonomura *et al.*, 2004) [12].

Spreading type mutant

Spreading type mutants were observed in ICGS-76. These were characterized by their dark green colour and excessive vegetative growth.

Table 1: Frequency of chlorophyll and viable mutants in M_2 generation of two groundnut varieties

Dose	ICGS-76			Rajashree			Total		
	No. of M2 Seedlings	No. of Mutants	Mutation Frequency	No. of M2 Seedlings	No. of Mutants	Mutation Frequency	No. of M2 Seedlings	No. of Mutants	Mutation Frequency
Control	300			300			600		
0.2%, NaN_3	300	6	2.00	300	-	-	600	6	1.00
0.4%, NaN_3	300	3	1.00	300	2	0.67	600	5	0.83
0.6%, NaN_3	300	7	2.33	300	2	0.67	600	9	1.50

Fig 1: Chlorophyll mutant viridis

Fig 2: Chlorophyll mutant variegated

Fig 3: Dwarf fertile mutant

Fig 4: Dwarf sterile mutant

Fig 5: Spreading type mutant

Mutagenic effectiveness and efficiency

Mutagenic effectiveness is a measure of frequency of mutations induced by unit dose of mutagen, whereas mutagenic efficiency is indicative of proportion of mutations as against undesirable biological effects such as gross chromosomal aberrations, lethality and sterility (Konzak *et al*., 1965) [8].

Mutagenic effectiveness

In the present study the in general mutagenic effectiveness was found to be highest for 0.2% NaN_3 (Table.2) indicating that mutagenic effectiveness was higher at lower dose. This agrees with the results obtained by Siddiq and Swaminathan (1968) [16], Prasad (1972) [13] and Satpute and Kothekar (1994) [14] in different plant systems. The greater effectiveness at lower doses of mutagens was due to the fact that biological damage increases with increasing doses (Konzak *et al*., 1965) [8].

The data presented in table.4 indicated that the effectiveness of NaN_3 and response of varieties was varying. In variety ICGS-76, 0.2% NaN_3 concentration was most effective while in Rajashree variety, 0.4% NaN_3 showed highest effectiveness value.

Mutagenic efficiency

Lethality and injury are indication of the sensitivity of a genotype to a mutagenic dose (Gaul, 1958) [4]. In the present study over all highest mutagenic efficiency in terms of both lethality and injury can be seen for 0.2% NaN_3 concentration (Table.3). El-Araqi *et al*., (1996) [3] reported chromosomes are damaged by induced mutation. Konzak *et al*., (1965) [8] reported treatments with lower concentration for longer period always result in a higher frequency of mutations and hence the mutagenic efficiency was greater in lower concentration.

The varietal differences in the mutagenic effectiveness and efficiency can be seen in Table 4. In variety ICGS-76, mutagenic efficiency in terms of both lethality and injury is highest at 0.2% NaN_3 concentration. In variety Rajashree, mutagenic efficiency in terms of both lethality and injury is highest at 0.4% NaN_3 Among the two varieties studied ICGS-76 showed higher mutagenic effectiveness as well as mutagenic efficiency when expressed in terms of both lethality and injury.

Table 2: Mutagenic effectiveness of NaN_3 in two groundnut genotypes

Treatment	Mutation Frequency			Mutagenic Effectiveness
	ICGS-76	Rajashree	Mean	
0.2% NaN_3	2	-	1	5.00
0.4% NaN_3	1	0.67	0.84	2.10
0.6% NaN_3	2.33	0.67	1.5	2.50
Mean	1.78	0.45		

Table 3: Mutagenic efficiency of NaN_3 in two groundnut genotypes

Treatment	Lethality			Injury			Mutagenic efficiency	
	ICGS-76	Rajashree	Mean	ICGS-76	Rajashree	Mean	Lethality	Injury
0.2% NaN_3	12.99	33.1	23.05	4.54	9.68	7.11	0.09	0.28
0.4% NaN_3	14.44	26.55	20.50	11.9	7.51	9.71	0.04	0.09
0.6% NaN_3	16.24	27.59	21.92	9.3	2.69	6.00	0.07	0.25

Table 4: Varietal differences in the Mutagenic effectiveness and efficiency of NaN_3 in two groundnut genotypes

Variety	Dose (%)	Lethality (L) (%)	Injury (I) (%)	Mutation Frequency	Effectiveness	Efficiency	
						Lethality	Injury
ICGS-76	0.2% NaN_3	13.00	4.55	2.00	10.00	0.15	0.44
	0.4% NaN_3	14.44	11.90	1.00	2.50	0.07	0.08
	0.6% NaN_3	16.25	9.31	2.33	3.88	0.14	0.25
Mean		14.56	8.59				
Rajashree	0.2% NaN_3	33.10	9.68	-	-	-	-
	0.4% NaN_3	26.55	7.51	0.67	1.67	0.03	0.09
	0.6% NaN_3	27.59	2.69	0.67	1.11	0.02	0.25
Mean		29.08	6.63				

Conclusion

The present investigation provided insights into the mutagenic effects of Sodium Azide on groundnut varieties. It demonstrated that the choice of mutagen dose plays a critical role in inducing mutations, and different groundnut varieties may respond differently to mutagenic treatments. Effectiveness and efficiency are both crucial factors in determining the usefulness of any mutagen. In the present study 0.2% NaN_3 was found to be most effective as well as most efficient dose. These findings can be valuable for the development of groundnut varieties with improved traits and increased genetic diversity, contributing to the enhancement of the groundnut industry. Further studies and breeding programs may build upon these results to harness the potential benefits of mutation breeding in groundnut cultivation.

References

1. Blixit S. Quantitative studies of induced mutations in Peas. V. Chlorophyll mutations. Agric. Hortic. Genet. 1961;19:402-47.
2. Duncan WC, McCloud DE, McGraw RL, Boote KJ. Physiological aspects of peanut improvement. Crop Sci. 1978;18:1015-1020.
3. El Araqi KT, Gilot-Delhalle J, Moutschen-Dahmen J, Moutschen-Dahmen M. Evaluation of Chromosome Damage due to a Nitronaphtofuran Derivative; R7372 on the Plant Nigella damascene. Cytologia. 1996;61(1):1-6.
4. Gaul H. Present aspects of induced mutations in plant breeding. Euphytica. 1958;7:275-289.
5. Gaul H. Mutation in plant breeding. Rad. Bot. 1964;4:155-232.
6. Goyal S, Wani MR, Khan. Frequency and Spectrum of Chlorophyll Mutations Induced by Single and Combination Treatments of Gamma Rays and EMS in Urdbean. Asian J Biol. Sci. 2019;12:156-163.
7. Gustafsson A. The mutation system of the chlorophyll apparatus. Lunds. Univ. Arrsr. N.F. Avd. 1940;30:1-40.
8. Konzak CF, Nilan RA, Wagner J, Foster RJ. Efficient chemical mutagenesis. The use of induced mutagenesis in plant breeding. Rad. Bot. 1965;5:49-70.
9. Niknejad M, Konzak CF, Donaldson E, Wickham I, Nilan RA, Sander C. Azide, a potent new mutagen near non-additivity of mutation yields from post-treatment combinations of barley with N-methyl-N' - Nitroso urea. Agron. Abst; c1972. p. 27.
10. Nilan RA, Konzak CF. Increasing the efficiency of mutation breeding. Mutation and plant breeding. NASNRC. 1961;891:931-460.
11. Nilan RA, E.G. Sideris, A. Kleinhofs, C. Sander, C.F. Konzak. Azide, a potent mutagen. Mut. Res. 1973;17:142-144.
12. Nonomura KI, Nakano M, Murata K, Miyoshi K, Eiguchi M, *et al*. An insertional mutation in the rice PAIR2 gene, the ortholog of Arabidopsis ASY1, results in a defect in homologous chromosome pairing during meiosis. Mol. Genet. Genomics. 2004;271:121-129.
13. Prasad MVR. A comparison of mutagenic effectiveness and efficiency of gamma rays, EMS, NMU and NG. Indian J. Genet. 1972;32(3):360-367.
14. Satpute RA, Kothekar VS. Mutagenic efficiency and effectiveness in safflower. J Nuclear. Agric. Biol. 1996;25(4):230-234.
15. Sharma A, Sharma P, Choudhary S, Sharma M, Kumawat KC, Khinchi SK. Studies on biology of Caryedon serratus (Olivier) on groundnut (*Arachis hypogaea* L.). J Pharm. Innov. 2021;10(12):3005-3008.
16. Siddiq EA, Swaminathan MS. Enhanced mutation induction and recovery caused by NG in Oryza sativa L. Indian J Genet. 1968;28:297-300.
17. Swaminathan MS. A comparison of mutation induction in diploid and polyploids. The use of induced mutation in plant breeding. Rad. Bot. 1964;5:619.
18. Swaminathan MS, H.K. Jain. Food legumes in Indian agriculture. In: Nutritional improvement of food legumes by breeding. Roma, S. ed., sponsored by protein advisory group U.N., Roma; c1973. p. 69-82.