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Effect of Chemically induced Mutation on Yield Contributing and Seed quality Attribute in Pea (*Pisum sativum* L.)

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ABSTRACT: Pea (*Pisum sativum* L.) is one of the important pulse crop in India, having chromosome number is 2n=24 and family is Leguminaceae. Peas are a valuable vegetable for vegetarians and fruits and seeds, both green and mature, are rich in starch, proteins. Mutation is the ultimate source creating variation and Breeder always want wider variability among population. Mutation breeding can improve the genotype. For improvement in the population variability is required, which can be created by various methods like hybridization, soma clonal variation, polyploidy. Mutation is also play an important role in creating variability. The present study was carried out to determine effect of chemically induced mutation on yield contributing and seed quality attribute in Pea (*Pisum sativum* L.) in M_1 generation. Seed were pre-soaked in distilled water for 6hrs and later treated with different concentration w/v (0.1, 0.15, 0.20, 0.25) of Sodium Azide and Hydroxyl Amine respectively for 6hrs. The experiment was planted Randomized block design. Observation in M_2 generation showed significant variation on the germination, root and shoot length, vigour index I and II, seedling dry weight, mortality, survival, primary secondary branches, days to first flowering, length of pod, number of pods per plant, seed test weight. Most of the parameters decreased with increase in concentration of Sodium Azide. The result of study showed that Sodium Azide 0.25 and 0.20 are more effective to induce variability in population. A significantly was observed effects on plant height, germination, days to first flowering and Days to first flowering. Sodium Azide is more effective as compared to the Hydroxyl Amine in inducing genetic variability.

Keywords: Pea, Mutant, Sodium Azide, Hydroxyl Amine, S10, Variability.

INTRODUCTION

Pea (*Pisum sativum* L.) pulse cum vegetable crop belongs to the family Leguminaceae, is a self-pollinated with 2n=24 chromosomes. Its Centre of origin is South Western Asia (Nepolian *et al.,* 2019). In India Pea covers 573 lakh hectares and produces 5823 million tonnes in India. Madhya Pradesh produces an average of 173.37 tonnes in 197.25 hac area and its yield is 879 (kgs/ha) in 2020-21 (Directorate of Pulses Development, Bhopal).

It's usually planted for its green pods, which contain immature seeds that can be cooked as a vegetable or eaten raw. Peas are a valuable vegetable for vegetarians as vegetable picked pea, canned, frozen and dehydrated or freeze and dried pea is consumed. Fruits and seeds, both green and mature, are rich in starch, proteins, oil, galactolipids, alkaloids, such as trigonelline and piplartine, as well as essential oils and soluble carbohydrates. Its high protein, starch, and absorbable supplement content, along with its low fibre content, makes it an excellent domestic animal feed. Natural or spontaneous mutations and artificial or forced mutations are the two forms of mutations. Natural mutations occur infrequently, thus artificial mutations are created. Induced mutagenesis with the use of mutagens is the most common way to increase genetic variety.

The most economical external input for farmers and a critical input for agricultural productivity is seed (Goyal *et al*., 2019 and Dhulgande *et al*. 2014). Ionizing radiation and chemical mutagens are among the mutagenic agents used to produce beneficial mutations at a high rate. The use of effective mutagens is crucial for successful mutant isolation. Hydroxyl Amine and Sodium Azide chemical mutagen used in plant research, causes single base changes with various mutation spectra.

MATERIALS AND METHOD

The study for the present investigation was conducted from October 2020 in the Crop Research Centre of Department (CRC), Department of Genetics and Plant Breeding, School of Agriculture, ITM University, Gwalior, M.P., India. The seeds of Pea (S 10) (*Pisum sativum* L.) were subjected to different treatment levels of mutagen *viz.* Sodium Azide (NaN_3) and Hydroxyl Amine (H_3NO). Treatment parameters were four different concentration (0-control) 0.1, 0.15, 0.20, 0.25 weight by volume. The solution of mutagen was prepared by dissolving 0.1, 0.15, 0.20, 0.25 gram of Sodium Azide and hydroxylamine in 100 ml of distil water respectively, and second to form 0.1, 0.15, 0.2, 0.25% weight by volume concentration of Sodium Azide and Hydroxyl Amine in 100 ml of distilled water, respectively and shaken to form 0.1, 0.15, 0.20, 0.25 per cent weight by volume concentration of Sodium Azide and Hydroxyl Amine respectively.

Before treatment seeds were pre-soaked in five different petri dishes containing distilled water for six hours at room temperature to enhance the effect of mutagen. There after five group of S 10 were subject to 4 (four) concentration (0.1, 0.15, 0.2, 0.25) of Sodium azide (NaN_3) and hydroxyl amine (H_3NO) solution and the control at room temperature 25°C for six hours. After treatment the seeds washed thoroughly with distilled water to remove the residual mutagens and air dried on filter paper (Ojua *et al.*, 2019). Germination of the seed started in about 7 to 15 days. 50 seeds of each treatment were sown immediately in plant to plant and row to row 10 × 30. The treated seed were laid out using Randomized Block Design (RBD) with three replications. Plants were properly irrigated and all the cultural practices were timely performed to evaluate the morphological and yield trait of S 10 with sodium azide and hydroxyl amine mutagen. The morphological trait studied include; Germination percentage, number of primary branches and secondary branches, days to first flowering, length of pod, number of pod per plant, yield per plot, mortality percentage, root length, shoot length, dry seed test weight, survival, mortality and seed test weight.

Data collected were subjected to Analysis of Variance using OP STAT (O.P. Sheoran Programmer, Computer Section, CCS HAU, Hissar) and significant mean were separated using t-test (One factor analysis). The data analyzed in RBD (One Factor) except laboratory like germination, root length, shoot length, dry seedling weight, Test seed weight. 32 plants were selected on the basis of significant change in the characters germination percentage, plant height and days to first flower and number of pod per plant. These selected plants were Suspected to be mutant for which the progeny generation of these plants was raised in the next year (October 2021). The number of plants raised in M_2 generation was 96 in three replications for each suspected mutant from M_1. The M_2 generation was planted with plan to plant and row to row distance as 30 × 10 cm. All the field practices were maintained as per the requirement to raise a good crop.

RESULT AND DISCUSSION

Mutants were characterized on morphological basis of plant type. Overall, 33 mutants were isolated the central matter in this mutation analysis concern the viable mutation may of which, whether they are morphological in character have potential value in plant breeding. The considerable rates of mutants were obtained with the mutagens Sodium Azide and Hydroxyl Amine. The frequency of the mutants with desirable, variable, morphological plants is present in the Table 3 and Fig. 1.

Mutants in M_1 generation (Table 1 & 2) were characterized with respect to the characters germination percent, plant height, days to first flower, Number of pod per plant. The characteristics of the identified mutant are discussed under the following heads. The frequency and saturation of mutations can be regulated by varying the mutagen dose. Mutagenic agents can induce different extensions of genomic lesions, ranging from base mutations to larger fragment insertions or deletions.

Table 1: Analysis of Variance for yield contributing attribute in M_1 Generation.

Treatment	Plant height		Primary branches		Secondary branches		Days to first flowering		No of pod per plant		Length of fruit		Mortality		Survival	
	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.
1	71.60	0.01	4.80	0.00	7.80	0.11	48.33	0.13	12.86	0.29	7.80	0.01	12.26	0.26	87.73	0.26
2	70.75	0.05	4.00	0.11	7.26	0.06	49.20	0.00	10.86	0.17	7.66	0.00	14.13	0.13	85.86	0.13
3	69.61	0.04	3.53	0.06	7.00	0.00	49.73	0.06	10.13	0.13	7.50	0.00	15.86	0.13	84.13	0.13
4	68.64	0.04	3.26	0.06	6.26	0.06	50.66	0.06	9.40	0.00	7.30	0.00	16.53	0.53	83.46	0.53
5	71.46	0.05	4.86	0.13	7.60	0.00	48.60	0.11	13.06	0.43	8.08	0.01	12.80	0.23	87.20	0.23
6	69.54	0.05	3.93	0.06	6.20	0.11	49.00	0.11	11.53	0.24	7.86	0.00	14.40	0.23	85.60	0.23
7	68.13	0.21	3.66	0.06	5.53	0.06	50.00	0.11	9.60	0.11	7.55	0.02	16.00	0.00	84.80	0.23
8	66.97	0.05	3.13	0.06	4.86	0.06	51.26	0.06	8.26	0.24	7.09	0.01	20.80	0.92	79.20	0.92
9	74.76	0.01	5.40	0.11	8.86	0.06	47.33	0.06	15.06	0.35	8.68	0.01	6.80	0.40	93.20	0.40
C.D.	0.24		0.15		0.23		0.26		0.68		0.04		1.13		1.11	
SE(m)	0.07		0.05		0.07		0.08		0.22		0.01		0.37		0.36	
SE(d)	0.11		0.07		0.10		0.12		0.32		0.01		0.53		0.52	
C.V.	0.19		2.16		1.95		0.31		3.51		0.29		4.52		0.74	

Sinjushin *et al*. (2022); Sinjushin *et al*. (2022); Parihar *et al*. (2022); and Giri *et al*. (2010); in Pea recorded that, the gamma rays caused more viable mutations followed by combination and EMS treatments. Sachin and Shama (2022); Savant *et al*. (2016) in Pea recorded that; frequency of viable mutants was higher in treatments with EMS than with gamma rays.

The data for plant height depicted that the plant height

taken in M_1 generation for the suspected mutant was comparable to the plant height emergence in the M_2 generation as well. In M_1 generation the selected plant at 64.6 days after sowing and similarly in M_2 generation of same treatment the plant height emergence took place at 64.5 days. The mother plant S10 plant height was 74.6 days. This shows an increase in the days to plant height in the suspected mutant. Umavathi and Mullainathan (2018); Sangle and Kothekar (2013).

The data for pod per plant depicted that the no of days taken in M_1 generation for the suspected mutant was comparable to the no. of pod per plant emergence in the M_2 generation as well. In M_1 generation the selected pod per plant was 6 pods and similarly in M_2 generation of same treatment the no. of pod per plant emergence took place at 6 pods. The mother plant S10 no of pod per plant 15. This shows increase in the days to pod per plant in the suspected mutant.

The data for days to first flowering depicted that the number of days taken for flowering in M_1 generation for the suspected mutant was comparable to the days to first flower emergence in the M_2 generation as well. In M_1 generation the selected plant flower at 54 days after sowing and similarly in M_2 generation of same treatment the first flower emergence took place 53 days. The mother plant S10 flower 47 days, this shown an increase in the days to first flowering in the suspected mutant.

The data for Germination depicted that the Germination taken in the M_1 generation for the suspected mutant was comparable to the germination emergence in the M_2 generation as well. InM_1 generation the selected plant germination at 64 and similarly in M_2 generation of same treatment the Germination emergence took place at 63 percent. The mother plant S10 Germination 85 Percentage. This shows increase in the days to germination in the suspected mutant. Vasisth *et al*. (2022); Jyothsna *et al*. (2022); Das *et al*. (2021); Sharma and Gautam (2019); Kalapchieva *et al*. (2021); Kirtane (2014) and Ariraman *et al*., (2014).

Table 2: Analysis of Variance for seed quality attribute in M_1 Generation.

Treatment	Germination		Root		Shoot		Dry seed		Vigour Index I		Vigour Index II		Seed test weight	
	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.
1	82.13	0.13	14.30	0.04	8.88	0.01	0.556	0.00	1,904.93	1.36	45.63	0.07	14.307	0.044
2	80.33	0.26	14.02	0.04	8.75	0.00	0.553	0.00	1,829.97	3.01	44.46	0.15	14.027	0.040
3	78.86	0.35	13.75	0.01	8.46	0.01	0.552	0.00	1,751.89	8.08	43.56	0.23	13.753	0.017
4	76.26	0.52	13.27	0.00	8.32	0.02	0.551	0.00	1,646.85	11.69	42.03	0.39	13.273	0.006
5	83.00	0.41	14.49	0.01	9.06	0.02	0.559	0.00	1,955.46	8.59	46.37	0.23	14.493	0.014
6	80.53	0.13	14.14	0.02	8.98	0.03	0.554	0.00	1,862.45	3.25	44.64	0.06	14.147	0.027
7	75.60	0.20	13.42	0.02	7.89	0.03	0.551	0.00	1,611.30	7.66	41.69	0.09	13.420	0.023
8	69.20	0.80	12.42	0.01	7.20	0.02	0.548	0.00	1,358.68	18.29	37.91	0.45	12.427	0.013
9	86.93	0.70	18.49	0.04	9.69	0.02	0.569	0.00	2,450.29	16.14	49.46	0.53	18.493	0.040
C.D.	1.36		0.08		0.07		0.002		30.78		0.88		0.084	
SE(m)	0.45		0.02		0.02		0.001		10.28		0.29		0.028	
SE(d)	0.64		0.04		0.03		0.001		14.54		0.42		0.040	
C.V.	0.99		0.34		0.51		0.239		0.97		1.17		0.342	

Table 3: Comparison studies of M_1 and M_2 Generations with Control.

Character	Mutagen	Dose	Variety	Control	M_1	M_2
Germination(%)	S.A	0.25	S10	85	64	63
Plantheight(cm)	S.A	0.25	S10	74.6	64.6	64.5
Plantheight(cm)	S.A	0.20	S10	75	65.3	65.1
Daystofirstflowering	S.A	0.25	S10	47	54	53.6
No. ofpodperplant	S.A	0.20	S10	14	6	6

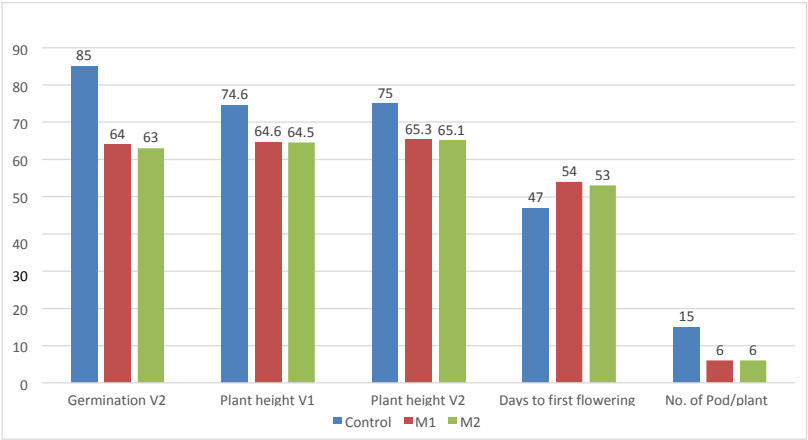

Fig. 1. Comparative representation of M_1 and M_2 Generations with Control.

CONCLUSION

The variation in the morphological features in the M_1 mutant, S10 is an indication that Sodium Azide is more effective as compared to the hydroxyl amine in inducing genetic variability in the quantitative trades generally, differences in concentration of the mutagen significantly affected most of the parameters evaluated in the given varieties. The result of study showed that Sodium Azide significant effect on plant height, germination and days to first flowering and Number of pod per plant. Therefore, induce mutation is highly effective in creating variability of desirable trade in Pea for further reading program.

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Conflict of Interest. None.

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