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COMPATIBILITY STUDY OF SODIUM AZIDE INDUCED MACROMUTANTS OF CAPSICUM ANNUUM L. VAR. PUSA JWALA.

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Abstract:
Capcicum annuum L. (Chilli) is an important cash crop in India and is grown for its pungent fruits which gives pungency to the food. The pungency is due to the active principle 'Capsaicin' contained in the skin and the septa of the fruit. Species C. *annuum* L. and *C. frutescence* L. (2n=24) are the two principal species grown in India and the varieties of the *C. annuum* constitutes the chief source of the dry chili of commercial use. Information relating to the occurrence of mutations either induced or spontaneous in species of *C. annuum* is vast. Genetic phenotypic variation in relation to yield components by using different mutagenic agents like EMS, NMU, MMS and gamma-rays were studied by several workers. However, information on S.A (Sodium azide) induced mutations in *Capsicum annuum* is quite meagre. This species attracted considerable attention because of its economic importance, mainly on the commercial aspects. Therefore, it is planned to subject *Capsicum annuum* L. var *pusa jwala* to experimental mutagenesis and assessed five macromutants and their compatibility with siblings and controls were discussed in the present paper.
Key Words: *Capsicum*, Compatibility, Macro mutant, Mutagen, Sodium azide,

Introduction:

Induced mutation is an important method in crop improvement in order to supplement existing germplasm and to improve cultivar for certain specific traits. Mutations cause's genetic alterations in DNA to gross changes in chromosome number and structure. Ever since the discoveries of mutagenic potential of x-rays by Muller (1927) and Stadler (1928), the possibility of obtaining very high mutation rates by artificial means have been realized by many plant breeders.

The usefulness of any mutagen in plant-breeding depend not only its mutagenic effectiveness, but also on its mutagenic efficiency. Efficient mutagen is the production of desirable changes free from associations with undesirable changes. However, the most effective agent or treatment may not be the most efficient. Konzak et. al. (1965) noted the apparent mutagenic effectiveness and efficiency of any two agents may differ in different organisms and tissues because of the comparable test conditions may not permit the expressions of the true potential of the agents.

In order to induce genetic variability and utilize useful mutation for efficient plant breeding, the systematic study of induced viable morphological mutations is the most dependable index; various methods have been made to determine the most effective and efficient mutagens and treatments for the induction of beneficial mutation in various crop plants.

Sodium azide (S.A) is mutagenic in several plant systems. It is also an inhibitor of oxidative phosphorylation and interferes with the mechanism of chromosome repair (Vig & Reenee Sung, 1981). It can be metabolized by plants into a mutagen. However there is no conclusive evidence implicating its role in cell metabolism.

Material and Methods:

The experimental material comprises *Capsicum annuum* L. Var. *pusa jwala* obtained from Ankur Seed Ltd. Company, Nagpur. It is a diploid, true breeding conformed, the plants were selfed before using for experiment. The chemical mutagen used in the present experiment was Sodium Azide (S.A). Stock solution of 1% sodium azide (S.A.) was prepared with phosphate buffer (pH 7) and were diluted to 0.1, 0.2, 0.3 per cent solutions as required. The seeds were pre-soaked in glass distilled water (pH 7) for 8 hr. Water was drained and excess water was blotted off, seeds were transformed to S.A. solution. To facilitate uniform absorption, large quantities of solution of mutagens, approximately three times the volume of the seeds were used (Konzak et al 1965). About 100 pre-soaked seeds were used for each treatment. The treatments were for 6 hr. duration at each concentration of S.A in darkness at night, at room temperature 26.5^{o}C ± 1^{o}C.

During the treatment, the Petri-dishes containing the mutagen solutions and seeds were shaken well at intervals of 15 min. As a post treatment, the seeds were washed in running water for 1 hr. The washed seeds were blotted and sown in flower pots. Untreated seeds were also sown simultaneously to serve as controls for comparison.

Result and Discussion

Sodium Azide Induced Macro Mutants:

For the systematic study of macro-mutants, induced viable morphological mutations is most important index. The mutation affecting gross morphological changes in leaf, flower and fruit were screened as viable macro mutants.

In the present investigation sodium azide was found to be effective in inducing a broad spectrum of mutants. In almost all the treatments, morphological mutants were isolated. In the present investigation, five viable macro mutants were isolated and characterized on the basis of the part of the plant body affected and presented in Table. 1.

Table1. : Description of morphological variants / mutants of *Capsicum annuum* var *pusa jwala* in M_1 generation.

S.No.	Name of the mutant	Mutagen Source (S.A in %)	Phenotype of Macromutant	Total mutants Isolated in M_1
1.	Curly leaf (CL)	0.1	Plant slender and profusely branched, dark green, curled leaves, flowers few and small with high pollen sterility.	1
2.	Early flowering mutant (EF)	0.1	Plant was robust, profusely branched with big leafs, flowers normal. Plant flowered 16 days early compared to its siblings and control.	2
3.	Late flowering mutant (LF)	0.2 0.3	The plants showed stunted growth bearing short branches, leaves darks green, flowering 10-15 days delayed.	2
4.	High yielding mutant (HY)	0.2	Robust bearing numerous flowers consequently more fruits were pronounced compared to control.	1
5.	Broad tip fruit mutant (BF)	0.1	Plant was robust, profusely branched, flowers and fruits normal size. But fruit tip was blunt.	1

Compatibility study of macro mutants:
Twelve crosses (including reciprocal) were made between macro mutants and controls and between the mutants having desirable characters (high yielding and early flowering).Almost all crosses exhibited compatibility except control X curly leaf mutant combination. The selected combinations were, Control X Early flowering mutant (EF), Control X Late flowering mutant (LF), Control X High yielding mutant (HY), Control X Broad tip mutant (BF), control X Curly leaf mutant (CL) and High yielding X Early flowering mutant. Reciprocal crosses were also performed for all combinations.

Among these 12 combinations, a total of about 50 crosses were made. Care was taken for timing of the maturation of stigma of the emasculated female plant and pollen of the male plant. **F_1** hybrids were produced for the crosses of all the combinations except control X curly leaf mutant. A total of 53 F_1 hybrids were developed from 120 crosses. However, some of the hybrid fruits were dropped off sometime during development. Finally, only 30 hybrid fruits were attained maturity from ten combinations of crosses. Data on seed set / fruit, germination percent and survival rate of F_1 hybrid seeds of all the twelve combinations were collected. (Table 2.).

1. Control X Early flowering mutant (EF): Resulted into 60.00% successful crosses and 45.00% hybrid fruits attained maturity. Those in reverse combinations exhibited 50.00% and 40.00% respectively. Some hybrids that did not attain maturity dropped off some time during development. F_1 seed germination percent was 58.00; survival rate on 24^{th} day was 60.00% while in reciprocal, percent germination and survival rates were 62.00% and 60.00% respectively.

2. Control X Late flowering mutant (LF): 40.00% crosses were successful and 50.00% F_1 fruits attained maturity in both direct and reciprocal crosses. The mean seed value per F_1 hybrid, F_1 seed germination percent and survival rate on 24^{th} day were more or less equal in both direct and reciprocal crosses.

3. Control X High yielding mutant (HY): crosses success was 50.00% and F_1 hybrid maturity was 60.00% in both direct and reciprocal crosses. However, per cent germination of F_1 seed and survival rate on 24^{th} day of direct crosses was less compared to reciprocal crosses.

4. Control X Broad tip fruit mutant (BF): Broad tip fruit mutant was highly pollen fertile (85.00%). 70.00% of crosses were successful and 71.00% of F_1 hybrids attained maturity. While in reciprocal 60.00% of crosses were successful and 66.60% F_1 hybrids reached maturity. No. of seeds / F_1 hybrid was also significantly increased. F_1

seed germination and survival rate were also very high compared to other cross hybrids. However, in reciprocal crosses, percent germination and survival rate were normal.

5. Control X Curly leaf mutant (CL): Curly leaf mutant was highly pollen sterile (80.00%). Crosses were not success, flowers dropped off sometime after pollination. Exhibited complete incompatibility. Development of self pollinated fruits was normal. Yield was drastically reduced in this mutant. However, in reciprocal crosses, 30.00% crosses were success, but only one F_1 hybrid developed. This hybrid was also dropped off sometime during development.

6. High yielding mutant (HY) X Early flowering mutant (EF): 50.00% of crosses were successful in both direct and reciprocal crosses. 66.60% F_1 hybrids attained maturity in direct cross where as in reciprocal 80.00% F_1 seed germination percent and survival rate of reciprocal was higher compared to direct crosses.

Table. 2. Compatibility of macro mutants with control and percent germination and survival rate of F_1 hybrids of *Capsicum annuum* L. var *pusa jwala.*

Sr.	Hybridization (crosses)	Compatibility crosses	Successful (Out of 10)		Matured F_1 hybrid		Seed set / F_1 hybrid	% of F_1 seed germination	% of F_1 survival
			No.	%	No.	%			
1	Control X EF	√	6	60.00%	2	33.3	50±0.11	58.00	60.00
2	EF X Control	√	5	50.00%	2	40.00	45±0.66	62.00	60.00
3	Control X LF	√	4	40.00%	2	50.00	38±0.71	52.00	61.00
4	LF X Control	√	4	40.00%	2	50.00	36±0.61	51.00	60.00
5	Control X HY	√	5	50.00%	3	60.00	28±0.71	52.00	60.00
6	HY X Control	√	5	50.00%	3	60.00	39±0.11	58.00	68.00
7	Control X BF	√	7	70.00%	5	71.00	85±0.11	90.00	85.00
8	BF X Control	√	6	60.00%	4	66.60	50±0.74	45.00	40.00
9	Control X CL	X	-	-	-	-	-	-	-
10	CL X Control	√	1	10.00%	-	-	-	-	-
11	HY X EF	√	5	50.00%	3	66.66	31±0.71	40.00	60.00
12	EF X HY	√	5	50.00%	4	80.00	60±0.86	75.00	62.00

EF - Early flowering mutant LF -Later flowering mutant HY - High yielding mutant
BF - Broad tip fruit mutant
CL - Curly leaf mutant √-Compatible X-Incompatible √ - Partial

Compatibility studies help to assess the prepotency of parents in hybrid combination. A judicious choice of parents promotes in improvement process leading to a well planned hybridization programme. Since, it helps the breeder to estimate the nature of gene action for the expression of economically important trait. Identification of parents with desirable traits and cross combinations showing high desirable trait, effects which will facilitate commercial exploitation of heterosis, isolation of pure lines among the progenies of heterotic F_1s and to design efficient breeding programme for crop improvement. Heterosis breeding has been useful in alleviating the food problem of the globe. This technique is well established in cross pollinated crop. Efforts are being strengthened to make the hybrid technology fulfil in self pollinated crops also. *Capsicum annuum* is one of the most important self pollinated tropical and subtropical commercial vegetable crops. So a study on hybrid breeding in chillies stands for beneficial. Therefore, the present study was under taken to identify good combining parents and heterotic crosses of mutants for important qualitative and pollen fertility characters.

In the present study, 99.00% compatibility was observed in all cross combinations except in curly leaf mutant. This mutant was high pollen sterile. Crosses were not success with control, both in direct as well as reciprocal crosses. But fruit formation was observed with self fertilization. Successful hybridization between the mutants and mutants with control revealed a high degree of compatibility between crosses (Subba Rao, 2003). Pollen viability test in the mutant supports a high degree of genetic incompatibility between them. The present investigation supports this, the low pollen viability might be the cause for genetic incompatibility in curly leaf mutant.

Similar results were also reported in *Capsicum* by Singh & Chowdhary (2003).

In overall some of the hybrids were dropped off sometime during development. As the endosperm is the chief source of nourishment for the developing embryo, the failure of cross may be attributed to some kind of physiological imbalance between the embryo and the endosperm. Copper and Brink (1942) termed this somatoplastic sterility, such embryo abortion may be the cause for the pre mature dropping of hybrid *Capsicum* in the present investigation.

The F_1 hybrid of Control X Broad tip fruit mutant exhibited positive heterosis over parent for F_1 seed germination and survival rate after a good scope for heterosis breeding in chilies. Thus the present study reveals that there is an ample scope for exploitation of hybrid vigour for commercial production as well as for isolation of pure lines among the progenies of heterotic crosses. Similar work was also carried by Khairwal & Sultan Singh (1999) and Subba Rao (2003) in pearl millet and Aniel Kumar et.al, (2006) in *Capsicum annuum*.

Conclusion

In the present investigation, high spectrum of macromutants were induced in 0.1% dose however, desirable macromutants (high yielding & early flowering) were isolated in 0.2%. So in the present study the most effective dose was 0.1% and the efficient dose was 0.2% for *Capsicum annuum* var. *pusa jwala*. These morphological mutants were utilised as indicators to their corresponding mutagenic (sodium azide) efficiency and also serves as good experimental material to investigate the genetics of floral, foliar and fruit morphogenesis and is useful for further improvement of the present taxon.

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