"QUANTITATIVE AND QUALITATIVE ASSESSMENT IN WILD CHICKPEA AND ITS INDUCED MUTANT"

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ABSTRACT

The chickpea is the third important legume crop of the world. The wild species of Cicer can be used in the cultivated chickpea breeding programme as a natural resource due to some important traits. Some of the undesirable traits of the wild species constraints its utilization in improvement programme and the crossability barriers in interspecific crossbreeding as well. Mutagenesis is a useful method to bring the desirable traits in the genome and elimination of undesirable traits. The induced mutants with suitable and desirable traits could be used indirectly in the breeding programme. The numbers of chemical and physical mutagenic agent are used in the mutagenesis. EMS and gamma rays are important chemical and physical mutagenic agents widely used to induce the mutation in the various plants species.

KEYWORDS- *Wild chickpea, M_3 Generation, EMS, Gamma radiation, Mutagenic agent.*

INTRODUCTION

Chickpea (*Cicer arietium*) is identified as cool season food legume with worldwide cultivation and India is a largest producer (Muehlbauer, 1993, Gebisa *et al.*, 2000). The genetic variation in chickpea has been tapped at large in the conventional breeding programme, narrowed the genetic variation base (Wani and Anis, 2008). The rotational cropping pattern with legume crop improves the soil fertility (Davies *et al.*,1985). Mutagens can be used to induce variability in plant species. Mutagenesis is an efficient tool to induce genetic variability in the plant species in short period (Micke,. 1988)..Mutation breeding is useful method to improve the economically important traits and elimination of the undesirable gene from the elites lines (Lippert *et al.*,1964). Improvement of the breeding value of mutants could be achieved through the union of the different mutant genes in the same genome (Gottschalk, 1986). The success rate of crossing hybridization between cultivated and wild species of chickpea has been reported more than 75% when wild species used as female parent (Singh and Ocampo, 1997). The wild species of Cicer are unused due to crossability barrier (Gowda and Gaur 2004). Some undesirable characters has been mentioned in the wild species which constraints the use of wild species in chickpea breeding programs (Jaiswal *et al.*, 1986).The mutants with desirable traits could be used in the hybridization programme to transfer specific gene into the genome of the cultivar variety. The mutagenesis could create many mutants alleles with various degree of modification (Brown, 2003). A number of Chemical and physical mutagens are significantly used to

generate induced mutants which inturn, attains the significant position in genetic science. Genetic variation in genotypes are of vital importance to tap in the improvement and breeding programme of cultivated variety. The EMS as chemical mutagen and gamma radiation as physical mutagen have been reported as important mutagens utilized to increase mutation frequency in plants (Borkar and More, 2010).Mutation breeding can be applied to create wide spectrum of variation in wild germplasm and These mutants may be used indirectly in breeding programmes as useful parents. (Micke and Donini 1993).Wild germplasm could be an important source of genetic variation for improvement of cultivar variety (Croser *et al.*, 2003). *C. echinospermum* and *C. reticulatum* are commonly used in chickpea improvement programs (Berger *et al.*, 2004).

MATERIAL AND METHOD

The germplasm of wild chickpea *Cicer reticulatum* was procured from the ICRISAT, Patancheru, India. The healthy seeds were treated with various concentration of ethyl methane sulphonate (EMS) such as 0.1/%,0.2,0.3 and 0.4% independently and were encoded as E2, E3,E4 and E5 respectively. The second set of healthy seeds were subjected to combined treatment of EMS and gamma rays. First the seeds were treated with EMS such as 0.1EMS +5kr Gamma rays,, 0.2EMS +10kr Gamma rays, 0.3EMS +15kr Gamma rays and 0.4EMS +20kr Gamma rays and encoded as E6, E7,E8,E9,respectively while untreated as Control C1.The treated Cicer seeds were sown to raise the M_1 generation to collect M_1 seed yield and sown to raise the M_2 generation and M_2 seed yield was collected to raise M_3 generation. The seeds alongwith the control were sown in the field following randomized block design (RBD) to raise M_3 generation in 3 replicates (Cochran and Cox, 1992). The seed-to-seed and row-to-row distance was maintained at 15 cm and 50 cm, respectively. Data for various phenological quantitative and qualitative profiles were collected at regular interval for computation to assess mean, standard error (SE), standard deviation (SD) and coefficient of variability (CV) using standard statistical procedure and ANOVA (Sukhatme and Amble, 1995).

RESULT AND DISCUSSION

The delayed germination was observed over control. 8.32 days as maximum mean period for germination in combined treatment while 6.10 day minimum mean period for germination in E_3 treatment were observed in present study. The mutagenic effect on various phenological character, flowering and fruiting were recorded at regular interval in M_3 generation and represented in the table 1.The maximum mean plant length 24.26 cm was observed in E_4 treatment and minimum 20.94 cm in E_6 treatment of M_3 generation. The reduction in height has been reported in *Solanum lycopersicum* treated with EMS and gamma radiation (Sikder *et al.*, 2013).The mean maximum stem length 3.96 cm was observed in E2 and minimum 2.94 cm was observed in E_8 treatment in M_3 generation during 20-40 days after sowing (DAS). The decrease in stem length was observed in the combination treatment.

The quantitative and qualitative effect of mutagenic treatment was observed in term of number of primary and secondary branches and length of primary and secondary branches.. The delayed branching was observed in the treatments E_4,E_5, E_7, E_8, E_9 over the control as reported previously in chickpea (Kamble and Petkar,2015).

The maximum number of primary branches i. e. 5.67 in E_5 treatment and 4.73 in E_7 treatment during 40 to 60 DAS. The variation in length of primary branches were observed in M_3 generation and 29.96 cm as maximum length in E_8 and 26.20 cm as minimum length in E_5 . The length of primary branches in all treatment was observed lower than the Control.

The plant height has been reported significantly higher in the chickpea treated with gamma rays and EMS (Wani and Anis,2008); in grasspea (Waghmare and Mehra, 2000). The increase in branching with increased number of fruits has been reported in *Brassica juncea* (Nayar and George, 1969). The minimum plant height was observed in the combined treatment of EMS and gamma rays in E_6 treatment than the control in the present assessment. The reduction in internodes length may be due to the reduction of cell length or the reduction of cell number (Weber and Gottschalk, 1973). The similar observation has been reported previously in Rhodes grass treated with gamma rays (Khan,1998), in *Solanum melanogena* (L.) treated with chemical mutagen (Alka *et al.*, 2007), in mungbean (Ansari *et al.*,1997).

The quantitative and qualitative variability was observed relative to the secondary branches in M_3 generation. The 5.07 as maximum number of secondary branches was observed in E_2 treatment and minimum 4.13 in E_4. The quantitative aspect of the secondary branches ranged between the control and the lowest concentration of EMS treatment was observed in M_3 generation during the course of present study. Qualitatively, 10.43 cm maximum length of secondary branches was observed in the E_2 treatment which was less than the length of secondary branches in the control (11.74 cm). The minimum length 6.93 cm was observed in the E_7 treatment.The primary and secondary branches were quantitatively recorded more in E_5, E_2 treatment respectively as compared to control in present study and in conformity with previous study in grasspea (Waghmare and Mehra, 2000), in chickpea (Kamble and Petkar, 2017).

The early flowering was observed in the treatment of lower concentration of EMS that is E_2 and E_3. The maximum number of flowers was found to be 7.82 in E_2 and minimum 6.82 in E_7 while in the control, it was 6.40 during the 90 to 110 DAS. The early pod formation was observed in all treatments in M_3 generation. The maximum 10.20 pods per plant were observed in E_2 treatment in M_3 generation while minimum 8.07 in E_4 and E_7 during 90 to 120 DAP. The mutagenic treatment was found to be non-significant with respect to two seeded pod and seed size in M_3 generation while the one seeded pod was observed significant.

The quantitative increase in pod per plant has been reported in chickpea treated with gamma rays (Wani and Anis,2008) , in grasspea treated with gamma rays and EMS in combination (Waghmare and Mehra,2000),in khesari (Singh and Chaturvedi,1990). An increase in flower, pod, seed has been reported in chickpea treated with EMS and gamma rays independently as well as in combination through the mutation breeding (Wani and Anis,2008). Mean number of capsule per plant and seed yield per plant has been reported as enhanced (Abo-Hegazi and Ragab, 1986). Increase in the seed yield has been reported in M_3 generation of *Lathyrus sativus* treated with gamma rays and EMS (Waghmare and Mehra, 2000) while seed yield has been reported as reduced in *Vigna mungo* (Singh *et al.*. 2000) No significant increase in number of seed per pod in mutant types has been reported in chickpea (Wani and Anis,2008, Kamble *et. al.,* 2015). The seed size was non-significant as reported in grasspea (Waghmare and Mehra,2000). The observations in present investigation revealed the conformity as reported in chickpea (Wani and Anis,2008). Mutagenic effectiveness has been reported as decreased with increased dose or concentration of mutagen in *Solanum lycopersicum* (Sikder *et al.*, 2013).The mutation in traits could be attributed to the mutation of pleiotropic gene or mutation of gene cluster or chromosomal arrangement as has been reported in chickpea (Wani and Anis, 2008).

CONCLUSION

The EMS and gamma radiation is chemical and physical mutagenic agent ,having potential to cause the mutation in the wild chickpea as revealed in the present study. It broadens the mutation spectrum in the wild germplasm. The wild species of the chickpea is

important on account of the resistance potential to various biotic and abiotic stresses from the viewpoint of improvement breeding programme. The useful traits in induced mutants may be harnessed for the betterment and improvement of the cultivated variety of chickpea.

The mutagenesis brings the variation in the wild species and may be useful in breeding programme. The E_2 treatment appeared as a fairly good among all. ANOVA for the treatments were observed significant ($p<0.05$). The comparative result on overall variability in M_3 generation was observed significant in present study.

REFERENCES

[1] Abo-Hegazi and Ragab, A. I .(1986), "A programme for improving oilseed in Egypt through Mutation breeding" In: Mutagenesis Basics and Applied (Eds. A B Prasad), Print House (India), Lucknow, 1986.

[2] Alka, M.Y.K. Ansari and Danish Shahab,(2007), " Effect of ethyl methane sulphonate (EMS) on seed germination, Plant height and pollen fertility of *Solanum melongena* L.," *India K. Applied and Pure Biso*. Vol. 22 (1): pp. 97-100.

[3] Ansari, B.A., Malik, A.J., Larik, A.S. and Ansari, K.A. (1997), " Interdependence of yield and its components in the hybrids of *Triticum aestivum* L.," *Pak. J. Agric. Agril. Engg. Vet. Sci.* 13 (2): pp. 19-20.

[4] Berger Jens, Neil C. Turner and Renee P. Buck (2004), "Wild and cultivated Cicer species- different evolutionary paths lead to different phonological strategies that can be exploited to broaden the adastation of chickpea (*C. arietinum* L.) in New directions for a diverse planet," Proc. of the 4th international Crop Science Congress Brisbane, Australia, 26.

[5] Borkar, A. T. and More, A. D. (2010), "Induced flower colour Mutation in *Phaseolus vulgaris* Linn. Thtrough physical and chemical mutagens," *Advances in Bioresearch* 1 (1): pp. 22-28.

[6] Brown, G. G. (2003), "The radish restore gene of Ogura cytoplasmic male sterility encoded a protein with multiple pentaricopettide repeats," *J. Plant* 35: pp. 262-272.

[7] Cochran, William G. and Cox, Gertrude M. (1992), Statistical Analysis 'Experimental Design' 2nd Edition , Wiley Classic Library Edition published 1992, A Wiley Interscience Publication John Wiley And Sons Inc, New York Chichester, Brisbane Toranto, Singapore. pp. 106-116.

[8] Croser, J. S., Ahmad, F., Clarke, H. J., Siddique, K. H. M. (2003), "Utilisation of wild Cicer in chickpea improvement - progress, constraints and prospects," *Aust. J. Agric. Res.* 54: pp. 429-444.

[9] Davies, D. R., Berry, G. J., Health, M. C. and Dawkins, T. C. K. (1985), In: Pea (Pisum sativum L.) (R.J. Summerfield and E.H. Roberts eds.) Williams Collins Sons and Co. Ltd. Landon. U.K.: pp. 147-198.

[10] Gebisa Ejeta, Randy A. Hautea, Josef-Franz Seitzer (2000), " System wide review of plant breeding methodologies," in the CGIAR, ICRISAT subpanel report, Patancheru ,India March 14-18, 2000. pp. 21-25.

[11] Gottschalk, W. (1986), Experimental mutagenesis in plant breeding. In: Mutagenesis Basics and Applied (Eds. A B Prasad), Print House (India), Lucknow.

[12] Gowda CLL, Gour PM (2004) Global scenario of chickpea research—present status and future thrusts. In: Ali M, Singh BB, Kumar S, Dhar V (eds) Pulses in new perspective. Indian Society of Pulses Research and Development, IIPR, Kanpur, pp 1–22

[13] Jaiswal, H. K., Singh, B. D., Singh, A. K. and Singh, R. M. (1986), "Introgression of genes for yield and yield traits from *C. reticulatum* into *C. arietinum*," *International Chickpea Newsletter* 14: pp. 5-8.

[14] Kamble, G.C., and Petkar, H. J. (2015), "Biotechnological Comparative Assessment of Mutagenic Agents on Morphological Traits and Phenology in Wild Chickpea," *Interational Journal of Science and Research* 4 (1) : pp.2604-2608.

[15] Kamble, G. C., Petkar, H. J.,Patil, A. S. (2015), "Biotechnological Study of Mutagenic Effect on Flowering and Fruiting Pattern in Wild Chickpea," *International Journal of Science and Research* 4(3): pp.-1398-1401

[16] Kamble, G.C., and Petkar, H. J. (2017), "Morphological Assessment Study In Wild Chickpea Treated With Mutagenic Agents", *Review of Research* 6(9):pp. 1-7

[17] Khan, I. A. (1998), "Determination of radio sensitivity in walnut (*Juglens regia*)," *J. Nuclear-Agriculture-and Biology* 27(3): pp. 218-219.

[18] Lippert, L. F., Berg, B. O., Cook, A. A. (1964), "Three variegated seedlings in the Pepper," *J. Hered.* 55:pp. 78-93.

[19] Micke, A. (1988), "Genetic improvement of grain legumes using induced mutations. An overview. In: Improvement of Grain Legume Production Using Induced Mutations," I. A. E. A., Vienna. 1-51: pp. 491-499.

[20] Micke A, Donini B (1993) Induced mutations. In: Hayward MD, Bosemark NO, Ramagosa I (eds) Plant breeding-principles and prospects. Chapman and Hall, London, pp 53–62.

[21] Muehlbauer, F. J. (1993), Food and grain legumes. In: J. Janick and J.E. Simon (eds.), New crops. Wiley, New York. pp. 256-265.

[22] Nayar, G. G. and George, K. P. (1969), "X-ray induced early flowering, appressed pod mutant in *Brassica juncea* coss," Radiation and radiomimetic substance in mutation breeding, Bombay: pp. 409-413.

[23] Sikder S, Biswas P, Hazra P, Akhtar S, Chattopadhyay A, Badigannavar AM, D'Souza SF (2013) Induction of mutation in tomato (*Solanum lycopersicum* L.) by gamma irradiation and EMS. *Indian J Genet* 73(4):392–399

[24] Singh, M. and Chaturvedi, S. N.(1990), "Improvement of yield and quality character of khesari da/ by use of mutagens," *Mysore J. of Agric. Sci*., 24: pp. 325-330, 1990.

[25] Singh, K. B. and Ocampo, B. (1997), "Exploitation of wild Cicer species for yield improvement in chickpea," *Theoretical and Applied Genetics.* 95 (3): pp. 418-423.

[26] Singh VP, Singh M, Lal JP (2000) Gamma ray and EMS induced genetic variability for quantita tive traits in urdbean (Vigna mungo (L.) Hepper). *Indian J Genet* 60(1):89–96).

[27] Sukhatme, P. V. and Amble, V.N. (1995), Statistical Method for Agricultural Workers, ICAR, New Delhi : pp.145-156.

[28] Waghmare, V. N. and Mehra, R. B. (2000), "Induced genetic variability for quantitative characters in grasspea (*Lathyrus sativus* L.)," *Indian J. Genet.* 60 (1): pp.81-87.

[29] Wani, A. A. and Anis, Mohammad(2008), "Gamma Ray- and EMS-Induced Bold-Seeded High-Yielding Mutants in Chickpea (*Cicer arietinum* L.)," *Turk. J. Biol*. 32: pp. 161-166.

[30] Weber, E. and Gottschalk, W. (1973), "Die Beziehungen Zwischen ZellgroBe and internodienlange beistrahleninduzierten" *Pisum – Mutanten. Beitr. Biol. Pfl.* 49: pp. 101-126.

Table 1: Effect of EMS and Gamma Ray on Phenological Charecters in M3 Generation

Treatment	Days for germination (Mean)	Stem Length (Mean)	Plant Length (Mean)	No of Primary Branches (Mean)	Length of Primary Branches (Mean) in cm	No of Secondary Branches (Mean)	Length of Secondary Branches (Mean) in cm	No of Flower (Mean)	No of Pods (Mean)	No of one seeded Pods (Mean)	No of two seeded Pods (Mean)	Size of seed (Mean) in gm
C_1	3.40	3.04	21.81	4.95	30.20	4.86	11.74	6.40	10.14	9.31	0.84	1.4679
E_2	6.14	3.96	24.05	4.95	27.82	5.07	10.43	7.82	10.20	8.86	1.35	1.4783
E_3	6.10	3.05	23.08	4.93	26.75	4.85	8.87	7.07	9.46	8.40	1.08	1.4738
E_4	7.21	3.24	24.26	5.42	26.44	4.13	7.08	7.35	8.07	6.94	1.14	1.5005
E_5	8.22	3.20	21.98	5.67	26.20	4.22	7.18	7.67	8.95	7.81	1.15	1.4655
E_6	6.15	2.95	20.94	5.08	27.08	4.76	7.45	7.41	9.54	8.27	1.28	1.5077
E_7	6.25	2.96	21.61	4.73	26.75	4.62	6.93	6.82	8.07	6.86	1.22	1.4648
E_8	8.26	2.94	22.40	5.47	29.96	4.15	9.48	7.07	8.14	7.06	1.09	1.4375
E_9	8.32	2.95	21.84	4.88	27.54	4.27	7.10	6.83	8.21	7.06	1.16	1.4438
F-Test	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	NonSig.	NonSig.
SE(m±)	0.016	0.016	0.038	0.010	0.019	0.011	0.065	0.020	0.005	0.004	0.002	0.0001
CD at 5%	0.049	0.049	0.114	0.031	0.056	0.032	0.194	0.060	0.015	0.012	---	----