



## Effect of Chemical Mutagen on Yield and Yield Attributing Traits in Chickpea (*Cicer arietinum* L.)

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### Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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### ABSTRACT

An experimental trail was conducted to differentiate the effect of sodium azide on yield and its attributes of chickpea. Overnight pre-soaked fresh seeds of ADBG-1, ICC-3020, ICCV-15112 genotypes were treated with various treatments of sodium azide (0.01%, 0.02%, 0.04%, 0.06%, 0.08% & control) for 06 hrs for the purpose of induction of mutation. The effect of sodium azide with unlike concentrations on yield and its attributing characters were studied in M1 generation of chickpea. Both laboratory readings and quantitative traits were recorded during experimentation. From the result, it was observed that all the laboratory readings exhibited reduction in their values upon increasing the concentrations of mutagen when compared to the control which was not treated. Whereas the efficacy of sodium azide on the average mean performance of quantitative traits on 03 chickpea genotypes exhibited that ICCV-15112 (0.04% SA) had higher seed yield and their attributing traits are Plant height, no. of secondary branches, biological yield and seed index.

*Keywords:* Chickpea; yield attributes; sodium azide and M1 generation.

### 1. INTRODUCTION

*Cicer arietinum* L. (chickpea) is a yearly grain legume crop, autogamous species with basic and

diploid chromosome number ranging 14 & 16. It stands at third place among all the legumes grown. It also provides a high-quality protein and its granular structures on hairs of leaves and

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Pods contain malic and oxalic acids. *k*-mer statistics show, chickpea genome is evaluated to be 738.09 Mb in size. It also indicates that 73.8% of the genome is apprehended in scaffolds [1]. There are two main types of chickpeas recognized in India, namely desi (90%) and kabuli (10%). Almost 90% of crop is grown in rainy season. Earliest record of chickpea is from the Hacilar site in Turkey, radiocarbon dated 5450 B.C. and rolled out worldwide <http://www.ikisan.com/tnbengalgramhistory.html> [2]. Estimates of average area of chickpea stands at 10.17 million hectares, with which it attained an average mean yield of 842 kilograms per Hectare and its productivity ranges to 11.35 million tonnes [3] [https://www.nfsm.gov.in/ReadyReckoner/CU1/Data\\_Jan2021](https://www.nfsm.gov.in/ReadyReckoner/CU1/Data_Jan2021).

Chemical mutagen sodium azide is a powerful mutagen in crop plants. Its mutagenicity is arbitrated through the production of an organic-metabolite of compound azide. Aforementioned metabolite induce through nucleus, in-cooperates the DNA, which leads to change in base pairs of a genome sequence also called point mutation. Dosage of mutagen application is a foremost contemplation in mutagenesis treatment. Overall, the increase in concentration of mutagen may cause higher level of deterioration. But as in the case of azide it is least dangerous and efficient chemical mutagen. Higher yields through mutations are attained at moderate M1 rates of sterility. The issues concerning metabolism, activity, cytotoxic and mutagenic effect of Sodium azide and comparison with other mutagens are presented [4].

## 2. MATERIALS AND METHODS

Current research consists of 03 diverse chickpea genotypes ADBG-1, ICC-3020, ICCV-15112 source from Agriculture research station, Adilabad and ICRISAT Telangana. The investigation was carried out at Genetics and Plant Breeding Research Farm of SHUATS, Naini, Prayagraj Uttar Pradesh in the course of Rabi 2020-21. Experiment was held in RBD (Randomized Block Design) with 03 replications. About 120 treated seeds were used for field experimentation and 90 seeds were used for laboratory experimentation of each genotype. The overnight pre-soaked seeds were treated with various concentrations namely 0.01%, 0.02%, 0.04%, 0.06%, 0.08% for 6 hours with sodium azide because the mutagen should have

to diffuse to the embryo through the seedcoat for effective mutagenesis, hence the long-term soaking of seeds may cause hydrolysis of sodium azide to hydrazoic acid is toxic and non-mutagenic. Therefore, each chickpea genotype has 05 treatment combinations with one control of each genotype thus 18 treated combinations were formed for the present study. The same process should be done for laboratory recordings. The distance between rows and plants was made 30×10 cm<sup>2</sup> spacing. Within each replication and plot, 05 plants were selected in randomized manner and labelled, elimination of plants in borders to decrease border effects. All the 11 quantitative traits were noted on 05 randomly picked plants excluding, days to 50% flowering and maturity where they were recorded from the overall average of the plot. Whereas in laboratory observations the characters were noted on 7<sup>th</sup>, 10<sup>th</sup> and 14<sup>th</sup> day count after treatment for germination percent, root, shoot length and seed vigour index. Treated seeds had kept in seed incubator chamber for effective germination.

## 3. RESULTS AND DISCUSSION

Current experiment focus on inducing genetic variability for improvement of crop by chemical mutagenesis in ADBG-1, ICC-3020, ICCV-15112 genotypes. Efficacy of sodium azide on the quantitative traits mainly Days to 50% Flowering, Days to Maturity, Plant Height (cm), No. of Primary branches per plant, No. of Secondary branches per plant, Total No. of Pods per plant, No. of Effective pods per plant, Biological Yield per plant (gm), Seed Index (gm), Harvest Index (%) and Seed Yield per plant (gm) were noted.

Analysis of Variance of eleven quantitative characters exhibited that the mean sum of squares with genotypes revealed higher significant variations for all traits studied at 1% level except for Days to 50% Flowering and Days to Maturity where it is at 5% level of significance. This indicates that the presence of greater level of difference among the genotypes. It also provides a great chance to improve genetic composition of the crop with direct selection of genotypes having the desirable traits. Whereas the degree of freedom (d.f) for replications stands at 02, for treatments it is at 17 and error is at 34. In replications and treatments the highest source of variation is showed for Harvest index (41.805\*\*) and lowest is noted for No. of Secondary branches per plant and No. of Primary branches per plant (0.254\*\* & 0.487\*\*).

Others with the same findings are [5, 6, 7, 8] in chickpea. Whereas mean performance of Days to Maturity and Days to 50% Flowering exhibits different results with different applications of sodium azide. While average mean data of the 1<sup>st</sup> flowering was observed at 48<sup>th</sup> day on untreated plants and average mean of Days to 50% Flowering were noted at 68 days [ADBG-1 (0.01% SA) & ICCV-15112 (0.04%)] to 75 days [ICCV-15112 (0.08% SA)] after sowing. Mean Days to maturity showed early maturity at 118 days [ICC-3020 (0.02% SA)] and delayed up to 125 days [ICCV-15112 (0.08%)]. Mean data on Plant Height (cm) ranged between 36.060 [ICC-3020 (0.08% SA)] to 43.930 [ICCV-15112 (0.04% SA)] and sodium azide, increase in concentration causes plants to exhibit decline in their growth and sub normality in few plants. Average mean of No. of Primary branches per plant varied from 1.660 [ICC-3020 (0.08% SA)] to 3.130 [ICC-3020 (0.02% SA)]. Maximum No. of Primary branches per plant was recorded in genotype ICC-3020 (0.02% SA). While No. of Secondary branches per plant were recorded as 3.130 [ICC-3020 (0.08% SA)] to 4.860 [ICCV-15112 (0.04% SA)]. Maximum No. of Secondary branches per plant were recorded in ICCV-15112 (0.04% SA). Therefore, as increase in concentration of mutagen both Primary and Secondary branches per plant show increase and decrease in their trend. Effect of mutagen on Total No. of Pods per plant and No. of Effective Pods per plant showed least average mean in ICC-3020 (0.08% SA) of 18.330 and 15.260 while the maximum mean for No. of Pods per plant and No. of Effective Pods per plant was recorded in genotype ICCV-15112 (0.02% SA) of 33.860 and 31.200 respectively. Whereas the Biological Yield per plant ranged from 5.460 [ICC-3020 (0.08% SA)] to 9.660 [ICCV-15112 (0.04% SA)], Harvest Index vary from 42.374 [ADBG-1 (CONTROL)] to 54.612 [ADBG-1 (0.02% SA)], its Seed Index differed from 20.000 [ADBG-1 (0.04% SA)] to 31.000 [ICCV-15112 (0.04% SA)] and Seed Yield per plant ranged from 2.460 [ICC-3020 (0.08% SA)] to 4.260 [ICCV-15112 (0.04% SA)].

From the above values the Biological Yield, Seed Index and Seed Yield per plant showed their highest values for ICCV-15112 (0.04% SA) which stands at 9.660, 31.000 & 4.260. Highest average mean of Harvest index was 54.612 which was noted in ADBG-1 (0.02% SA). Standard error (SE) of mean data is highest for Harvest Index (3.483) and least for No. Primary branches per plant (0.074) Whereas, comparing

with laboratory readings they showed, the increase in the concentrations of genotypes leads to clear cut decline in their values of Germination Percentage (99.480 to 33.330), Root Length (2.300 to 0.700), Shoot Length (4.000 to 1.200) and Seed Vigor Index (630.000 to 76.000) for 03 genotypes of 90 seeds each. Increase in concentration of mutagen is due to inhibitory effect of mutagens on germination or altered enzyme activity [9].

In current investigation, the quantitative characters and laboratory readings, were subjected to analysis and assessed the range of variability induced through sodium azide in 1<sup>st</sup> generation, to analyse the effect on yield and also to assess the relationship of yield and yield attributes in chickpea population. As anticipated, the difference among M1 population exhibited that the treated plants were having higher variability when they were compared to control in all the examined parameters shows negative and positive shifts. All the traits showed nonlinear distribution for all the 03 genotypes, they were increased and decreased, when there is an increase in concentration of sodium azide maximum depletion of quantitative characters were noted at ADBG-1, ICC-3020, ICCV-15112 (0.08% SA) concentrations due high level of mutagenicity. The reduction in quantitative characters in M1 generation were also reported by rice, [10] in an *Arabidopsis thaliana* and [11] in mung bean [12]. Consequently, the differences in the population treated was relatively higher than the control for the traits examined. No. of Primary and Secondary branches per plant exhibited positive and negative shifts in average mean value of chemical treatment in chickpea. With the concentration increase, the Days to 50% Flowering and Days to Maturity exhibited notable variations towards positive and negative trend, this results in enough variability in the population treated, and it also assists in the selection of late-flowering or early flowering plants. The Total No. of Pods per plants exhibited consecutive depletion with raise in mutagen concentration. Same reductions were also observed by [5] in chickpea, [13] in mustard and [14] in other plants. Mean data of Seed Yield per plant was in irregular manner in M1 generation of chickpea. Seed Index is a principal character used to calculate ability of yield in chickpea. In the current investigation, the Seed Index showed drop in trend from control except for ICCV-15112 genotype which showed irregular trend.

**Table 1. ANOVA among 03 genotypes for 11 quantitative traits in chickpea.**

<b>S.no</b>	<b>Source of Variations</b>	<b>Replications</b>	<b>Treatments</b>	<b>Error</b>
	<b>d.f</b>	<b>2</b>	<b>17</b>	<b>34</b>
1.	Days to 50% Flowering (DAS)	1.548	17.648 *	6.962
2.	Days to Maturity	0.955	15.853 *	7.459
3.	Plant Height (cm)	0.364	15.199 **	5.902
4.	No. of Primary branches/plant	0.086	0.254 **	0.016
5.	No. of Secondary branches/plant	0.001	0.487 **	0.054
6.	Total No. of Pods/plant	0.213	35.102 **	5.689
7.	No. of Effective Pods/plant	0.421	36.957 **	4.983
8.	Harvest Index (%)	5.182	41.805**	36.396
9.	Biological Yield/plant (g)	0.03	2.650 **	0.335
10.	Seed Index (100 seed wt.) (g)	0.216	31.037 **	4.67
11.	Seed Yield/plant (g)	0.02	0.644 **	0.119

\* Indicates significance at 5% level of significance &amp; \*\* Indicates significance at 1% level of significance

**Table 2. Mean performance for and 11 quantitative traits of 03 genotypes (5 conc. and 1 control) during Rabi 2020**

<b>Genotypes/traits</b>	<b>DF 50%</b>	<b>DM</b>	<b>PH</b>	<b>NPB</b>	<b>NSB</b>	<b>TNPP</b>	<b>NEPP</b>	<b>HI</b>	<b>BYLD</b>	<b>SI</b>	<b>SYLD</b>
<b>ADBG-1</b>											
<b>CONTROL</b>	70.000	121.000	40.800	2.260	4.200	29.200	24.660	42.374	6.900	28.100	2.900
<b>0.01% SA</b>	68.000	120.000	42.800	2.460	4.400	28.530	24.730	49.952	8.200	26.000	4.100
<b>0.02% SA</b>	72.000	124.000	41.930	2.460	4.260	32.200	27.660	54.612	7.200	21.720	3.930
<b>0.04% SA</b>	74.000	122.702	39.260	2.330	4.130	26.660	21.730	48.683	6.730	20.000	3.260
<b>0.06% SA</b>	71.000	120.000	39.530	2.330	4.200	27.860	24.400	47.958	7.400	27.000	3.530
<b>0.08% SA</b>	69.000	123.000	41.060	2.530	4.060	26.330	22.400	50.169	7.860	25.460	3.860
<b>ICC-3020</b>											
<b>CONTROL</b>	74.000	124.000	37.460	2.129	4.260	27.400	23.130	50.895	7.460	29.000	3.800
<b>0.01% SA</b>	71.000	121.000	40.130	2.330	4.600	29.060	25.400	49.089	8.000	31.000	3.930
<b>0.02% SA</b>	68.000	118.000	37.130	3.130	3.930	24.460	20.930	44.120	7.730	27.000	3.400
<b>0.04% SA</b>	74.000	124.000	38.260	2.261	4.260	27.330	22.530	48.335	7.600	22.320	3.600
<b>0.06% SA</b>	73.000	125.000	37.600	2.200	4.730	27.200	22.460	43.877	7.600	27.000	3.330
<b>0.08% SA</b>	75.000	123.000	36.060	1.660	3.130	18.330	15.260	45.387	5.460	23.000	2.460
<b>ICCV-15112</b>											

<b>Genotypes/traits</b>	<b>DF 50%</b>	<b>DM</b>	<b>PH</b>	<b>NPB</b>	<b>NSB</b>	<b>TNPP</b>	<b>NEPP</b>	<b>HI</b>	<b>BYLD</b>	<b>SI</b>	<b>SYLD</b>
<b>CONTROL</b>	69.000	118.166	39.400	2.200	3.600	28.460	25.530	49.854	7.729	27.000	3.860
<b>0.01% SA</b>	72.000	120.366	43.400	2.130	4.060	33.860	31.200	42.988	9.600	29.450	4.130
<b>0.02% SA</b>	70.000	124.000	39.130	2.330	4.330	28.330	25.060	44.918	8.061	30.060	3.600
<b>0.04% SA</b>	68.000	119.000	43.930	2.660	4.860	33.200	29.800	44.176	9.660	31.000	4.260
<b>0.06% SA</b>	71.000	123.000	37.600	2.130	4.400	27.130	23.600	42.892	7.930	24.000	3.400
<b>0.08% SA</b>	75.000	125.000	40.330	2.260	3.799	30.000	26.530	53.562	7.660	25.000	4.100
<b>Mean</b>	71.333	121.958	39.767	2.322	4.178	28.086	24.278	47.436	7.710	26.340	3.636
<b>S.E.</b>	1.523	1.577	1.403	<b>0.074</b>	0.134	1.377	1.289	<b>3.483</b>	0.334	1.248	0.200

**DF50%:** Days to 50 % Flowering (DAS), **DM:** Days to Maturity, **PH:** Plant Height, **NPB:** No. of Primary Branches per plant, **NSB:** No. of Secondary Branches per plant, **TNPP:** Total No. of Pods per plant, **NEPP:** No. of Effective Pods per plant, **HI:** Harvest Index, **BYLD:** Biological Yield per plant, **SI:** Seed Index (100 seed wt.), **SYLD:** Seed Yield per plant.

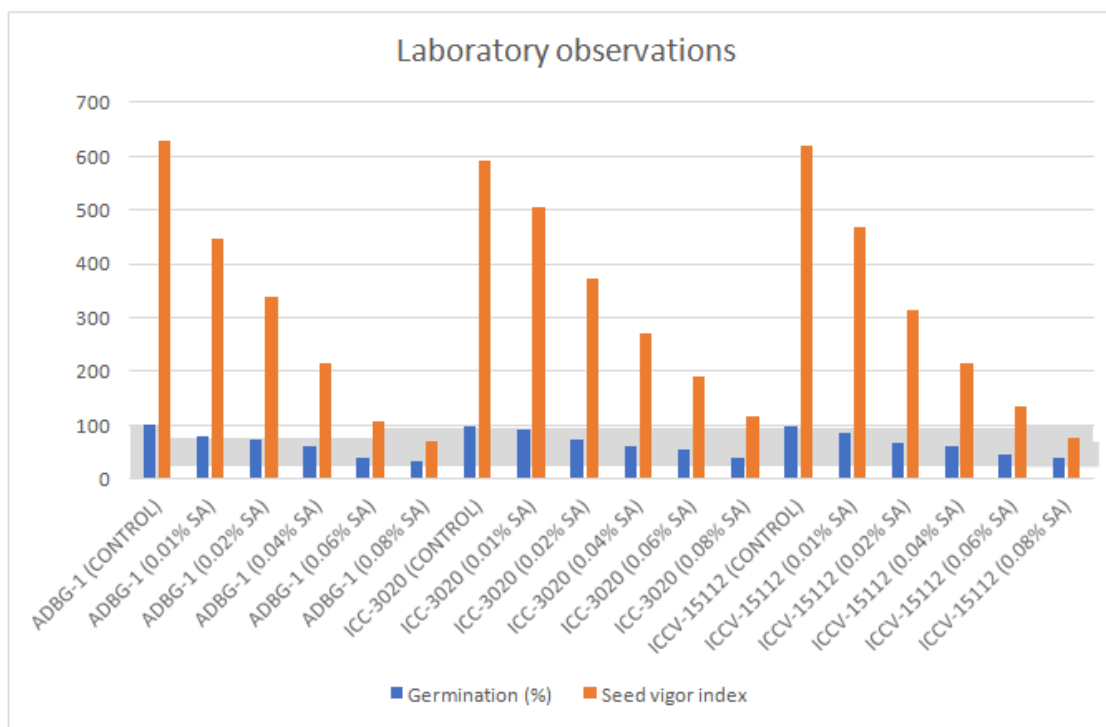


Fig. 1. Laboratory Observations (Germination Percentage & Seed Vigour Index) were based on 7<sup>th</sup>, 10<sup>th</sup> & 14<sup>th</sup> days after treatment on 90 seeds of 03 genotypes

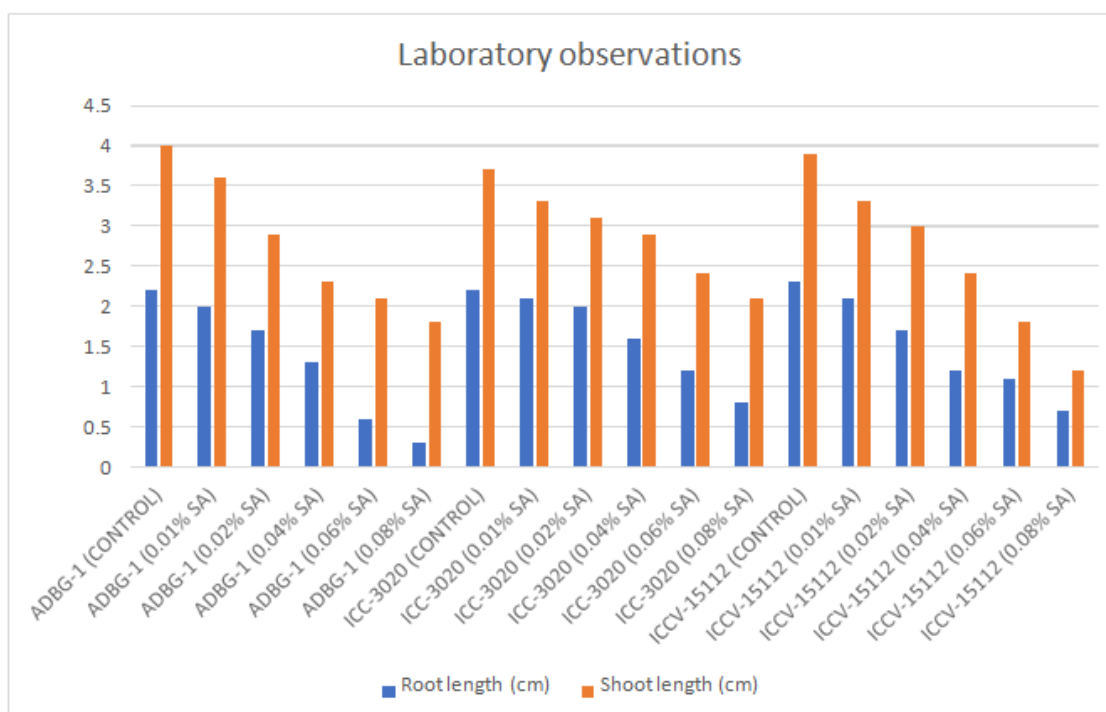


Fig. 2. Laboratory Observations (Root & Shoot length) were based on 7<sup>th</sup>, 10<sup>th</sup> & 14<sup>th</sup> days after treatment on 90 seeds of 03 genotypes

**Table 3. Genotypic (rg) & Phenotypic correlation (rp) coefficient Estimation between Yield and its attributetraits in 11 quantitative traits of 03 genotypes (5 conc. and 1 control).**

TRAITS	DF 50%	DM	PH	NPB	NSB	TNPP	NEPP	HI	BYLD	SI	SYLD
DF 50%	1	0.954**	-0.710**	-0.883**	-0.367**	-0.311*	-0.440**	0.698**	-0.683**	-0.754**	-0.484**
	1	0.4780***	-0.2405	-0.4549***	-0.2514	-0.2132	-0.1894	0.1599	-0.3352*	-0.21	-0.1773
DM		1	-0.553**	-0.611**	0.1173	-0.295*	-0.2664	0.838**	-0.509**	-0.638**	-0.303*
		1	-0.1715	-0.3304*	-0.0098	-0.0177	-0.2086	0.1764	-0.2377	-0.1957	-0.0679
PH			1	0.386**	0.551**	0.932**	0.941**	0.459**	0.897**	0.412**	0.970**
			1	0.1913	0.2079	0.4789***	0.6296***	0.0761	0.4596***	0.1851	0.458**
NPB				1	0.388**	0.2632	0.2659	0.343*	0.452**	0.229	0.512**
				1	0.3334*	0.2925*	0.2212	-0.0836	0.3459*	0.11	0.2246
NSB					1	0.669**	0.505**	-0.1862	0.636**	0.468**	0.556**
					1	0.4358***	0.4493***	-0.1929	0.4780***	0.2774*	0.2639
TNPP						1	0.932**	0.640**	0.907**	0.460**	0.925**
						1	0.6940***	-0.0323	0.5679***	0.2794*	0.475**
NEPP							1	0.442**	0.875**	0.521**	0.933**
							1	-0.0377	0.7231***	0.3781**	0.616**
HI								1	0.0652	-1.188**	0.361**
								1	-0.3427*	-0.1407	0.548**
BYLD									1	0.679**	0.957**
									1	0.5040***	0.593**
SI										1	0.375**
										1	0.350**
SYLD											1
											1

\* Indicates significance at 5% level of significance, \*\* Indicates significance at 1% level of significance & \*\*\* Indicates significance at 0.1% level of significance

The present experiment on genotypic correlation matrix analyzed that Seed Yield per plant was highly significant and positively associated with Plant Height (0.970\*\*), Biological Yield per plant (0.957\*\*), No. of Effective Pods per plant (0.933\*\*), Total No. of Pods per plant (0.925\*\*), No. of Secondary branches per plant (0.556\*\*), no. of Primary branches per plant (0.512\*\*), Seed Index (0.375\*\*) and Harvest Index (0.361\*\*). While Days to Maturity (-0.303\*) and Days to 50% Flowering (-0.484\*\*) shows lower significance and negative relation with Seed Yield per plant. Vice versa, at phenotypic level of correlation matrix the Seed Yield per plant had the highest significant and positive correlation with No. of Effective Pods per plant (0.616\*\*), Biological Yield per plant (0.593\*\*),

Harvest Index (0.548\*\*), Total No. of Pods (0.475\*\*), Plant Height (0.458\*\*) and Seed Index (0.350\*\*). While No. of Secondary branches per plant (0.263) and no. of Primary branches per plant (0.224) shows lower non significance and positive correlation to Seed Yield per plant at phenotypic level. Whereas Days to 50% Flowering (-0.1773) and Days to Maturity (-0.0679) shows the lowest non significance and negative correlation to Seed Yield at phenotypic level. The current correlation matrix results showed that the genotypic correlation was higher than the phenotypic correlation coefficient. Since, the interrelationships were strongly inherent and phenotypic correlation coefficient expression, appeared to be debilitated in some cases due to environmental impact thus selection based on phenotype may be effectual. The yield related traits showing positive and negative significant relation with Seed Yield per plant indicated that with Seed Yield per plant indicated that Seed Yield can be ameliorated by simultaneous selection for these characters. Hence, selection is based on phenotypic expression of traits. So, selection for traits showing positive significant genotypic and phenotypic correlation would be of higher use in indirect and direct selection for Seed Yield respectively. Research on the similar findings is seen in [15] wheat and [16] rice.

#### 4. CONCLUSION

All the genotypes with 18 different concentration combinations showed high levels of genetic variability. The genotype ICCV-15112 (0.04% SA) has highest seed yield and its yield attributing traits are Plant height, no. of secondary branches, biological yield and seed index during field experimentation. Vice versa, ADBG-1 (control) shown better performance when

compared to other treated and untreated genotypes in laboratory observations. Due to environmental effect the change in the observations were recorded in the field. Hence, improvement of this genotype concentration by contemplating to quantitative traits in upcoming generations. Present investigation shows the results, that variations in mutagen treatments had notable effect on all the quantitative traits studied. Induced Mutagenesis with polygenic variability has greater level of extent in improvement of chickpea by assimilating in conventional breeding techniques.

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#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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