

# Impact of hybridization and induced mutagenesis on variability of traits and resistance to *fusarium* wilt in Chickpea (*Cicer arietinum* L.)

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## Abstract

A high yielding and a late wilting variety Bheema was crossed to ICCV-10, also a high yielder but highly-resistant to wilt. The two parents are known to differ in respect of a single gene governing resistance to wilt. The F<sub>2</sub> seeds of cross between Bheema and ICCV-10 were subjected to gamma irradiation. Simultaneously, F<sub>2</sub> was also advanced to F<sub>3</sub> generation. Besides, Bheema was also subjected to irradiation to look to the possibility of developing high yielding and disease resistant mutants. The F<sub>2</sub>M<sub>1</sub> population proved to be highly potential in recovering higher frequency of high yielding and wilt resistant types than selfed (F<sub>3</sub>) and mutant (M<sub>2</sub>) populations.

**Key words:** Chickpea, *fusarium* wilt, variability, mutation, hybridization

## Introduction

Chickpea is a premier pulse crop of India. In backdrop of poor adaptability, conserved variability and susceptibility to biotic and abiotic factors the pulse crops have not been much successful and the chickpea is one of them. In view of this, efforts have been made to create variation in chickpea through hybridization [1, 2] and induced mutations to influence the productivity levels and to increase the resistance against fusarium wilt caused by *Fusarium oxysporum* f. sp. *ciceri*. Induced mutations were proved successful in many crops for the creation of wide genetic variability which was far reachable objective when wide hybridization leads to hybrid sterility or hybrid breakdown. These mutations were also found promising to correct the popular and high yielding cultivars which are lacking resistance to pest and diseases. Fusarium wilt is one of the serious diseases leading to major yield losses of chickpea. Many

fungicides are available in the market to control these diseases which are not cost effective. Considering economics of this crop developing cultivars with resistance to wilt is best solution for this problem. The reported data on fusarium wilt in chickpea indicated the existence of dominance for one of the two loci. The complete recessiveness of these loci will make the crop resistant. For this, mutation has profound impact to mutate the dominant locus into recessive condition.

## Materials and methods

In the present investigation, chickpea cultivars Bheema and ICCV-10 were used as susceptible and resistant sources for fusarium wilt reaction, respectively. Bheema is an excellent variety known for bold, golden coloured attractive seeds with good productivity. But it is susceptible to fusarium wilt. ICCV-10 is small seeded cultivar known to be resistant to fusarium wilt and diverse from Bheema with respect to productivity traits. To recombine the supplementary productivity traits of Bheema and ICCV-10 and also to transfer the resistance of ICCV-10, the hybridization between these two parents was carried out during *rabi* 2007-08. The F<sub>1</sub> seeds were harvested and were grown for generation advancement during *kharif* 2008 using off-season nursery. About 250 F<sub>2</sub> seeds harvested from F<sub>1</sub> plants were subjected to  $\gamma$  irradiation @ 60 kr during 2008 and remnant F<sub>2</sub> seeds were kept under proper storage conditions. Similarly, M<sub>1</sub> of Bheema obtained by subjecting 250 seeds of Bheema to  $\gamma$  irradiation (during *rabi* 2008-09) following the same dose was advanced to get M<sub>2</sub> of Bheema. And also un-irradiated F<sub>1</sub> plants which were grown in off-season nursery during *kharif* 2008 were advanced

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to get  $F_2$ . Thus, the experimental material comprising the  $F_2$   $M_1$ ,  $F_2$  and  $M_2$  of Bheema was generated. The experiment with these materials along with the parents was laid out at Dharwad during *rabi* 2008-09. The material was evaluated in an un-replicated trial for assessing the productivity and other component traits such as number of pods and seed weight. Using a part of seeds of all these generations evaluation was also made to assess for wilt reaction in wilt sick garden of ICRIAT simultaneously. Observation on number of pods per plant, 100 seed weight (g) as well as yield per plant (g) were recorded on 1500 plants in  $F_2$ ,  $M_1$  1200 plants in  $F_2$  and 1500 plants in  $M_1$  of Bheema. The data was subjected to variability analysis following standard statistical procedures to compute PCV, GCV, mean and range.

### Results and discussion

Host plant resistance to race 1 of wilt in chickpea is reported to be governed by three independent genes  $h_1$ ,  $h_2$  and  $h_3$  [2]. Recessiveness at the first two loci and partial dominance at the third locus confers resistance in the host plant. However the recessiveness at one and dominance at other locus would result in late wilting. Bheema, one of the parents used in present study is a late wilter. Hence, genetically it is presumed that it will be  $h_1 H_2 h_3$  or  $H_1 h_2 h_3$ . On the other hand ICCV-10 is the other parent used in the study is highly resistant to wilt. Its genetic constitution for wilt reaction locus is  $h_1 h_2 h_3$ . Both Bheema and ICCV-10 are high yielders known for their productivity potential. However, the traits contributing to the higher productivity in these two varieties are distinct. Hundred seed weight (32-35 g) is an important component of yield in Bheema while, higher pod number per plant is primary yield contributing trait in ICCV-10. Therefore, with the hypothesis that it should be possible to obtain optimum combination of distinct yield contributing traits with the possibility of recovering genotypes with still higher productivity through recombination breeding involving these two genotypes is proposed. At the same time this cross combination is also expected to help in transferring wilt resistant genes in segregants derived from this cross combination. It is therefore expected that it should be possible to recover high yielding and wilt resistant lines from this cross combination. Mutation is useful for generating variability in crops (cowpea, rice, and groundnut). Mutation in heterozygous population helps in releasing broadest spectrum of variability than mutation on homozygous population. The frequency of mutant derivatives with recessive homozygous condition is obviously higher

**Table 1.** Study on statistical parameters in chickpea breeding populations

Breeding population	Statistical estimates	No. of pods/plant	100-seed weight (g)	Seed yield/plant (g)
$M_2$ of Bheema	Mean	116.23	33.14	38.67
	PCV	36.96	18.92	36.51
	GCV	29.65	18.38	31.61
	Range	48-180	23.6-42.4	14-62
$F_2$ of Bheema x ICCV-10	Mean	126.42	22.5	33
	PCV	47.01	19.96	46.70
	GCV	38.58	19.35	42.61
	Range	35-330	10.6-39	4-89
$F_2M_1$ of Bheema x ICCV-10	Mean	158.83	22.53	38.80
	PCV	48.85	20.43	48.28
	GCV	43.89	19.85	41.08
	Range	17-405	15.6-35.8	7-91

when heterozygous genotype is subjected to mutation than when homozygous genotype was subjected to mutation. Considering this, heterozygous population of this cross ( $F_1$ ) was subjected to  $\gamma$  irradiation (60 kr) in order to improve the probability and prospects of realizing higher frequency of wilt resistant types. The similar attempts were made in cowpea by irradiation of heterozygous loci which gave the positive results [3].

These hypothetical considerations made on the bases of scientific reasoning have been realized. The  $F_2M_1$  population has shown higher mean value and higher range of expression for yield per plant as well as number of pods per plant and 100 seed weight. Similarly the percentage of wilt resistant segregants is 59.2 as against the percentage of resistant types in  $M_2$  of Bheema (29.21%) and  $F_2$  population (22.33%).

### Variability for productivity traits in segregating population

**Mean :** In respect of pods per plant the mean of  $F_2M_1$  (158.83) was much higher than  $F_2$  (126.42) and  $M_2$  (116.20). For seed weight mean of  $M_2$  (33.14g) was much higher than that of  $F_2$  (20.16g) and  $F_2M_1$  (22.50g). The mean seed yield per plant for  $F_2M_1$  and  $M_2$  (38.8g and 38.6g, respectively) were more or less same and slightly higher than  $F_2$  (33.8g). From this it may be concluded that  $F_2M_1$  was a better population than  $F_2$  or

M<sub>2</sub> since it showed high mean for seed yield as well as pods per plant.

**Variability:** The coefficient of variation (CV) values were computed to assess the extent of variation in three populations. For all the three characters F<sub>2</sub>M<sub>1</sub> showed the highest CV which is closely followed by that of F<sub>2</sub> in respect of seed yield and pods per plant. The CV of M<sub>2</sub> for seed yield and pods per plant was considerably less. However it was more or less same in respect of seed weight. Again on the basis of coefficient of variation, F<sub>2</sub>M<sub>1</sub> was a better population than either F<sub>2</sub> or M<sub>2</sub>.

**Range:** In respect of seed yield, the range of variation of F<sub>2</sub>M<sub>1</sub> and F<sub>2</sub> was comparable and high compared to that of M<sub>2</sub>. However, for number of pods per plant the range of expression of F<sub>2</sub>M<sub>1</sub> was much higher compared to F<sub>2</sub> and that of M<sub>2</sub> was much less. But the range of expression for seed weight of F<sub>2</sub> was highest followed by that of F<sub>2</sub>M<sub>1</sub> and M<sub>2</sub> while, upper range of expression for seed weight was highest in case of M<sub>2</sub>. On the basis of range of expression also F<sub>2</sub>M<sub>1</sub> again proved to be a better population than F<sub>2</sub> and M<sub>2</sub> population.

In chickpea, Singh *et al.* [4] studied association analysis of grain yield and its components following hybridization and a combination of hybridization and mutagenesis. The study revealed (1) insignificant positive correlation between all traits except 100 seed weight, which showed negative correlation with all other traits (in non-irradiated material), (2) Reduction of negative correlation of 100 seed weight particularly with seed yield/plant following irradiation and (3) Irradiation reduced various negative direct and indirect effects of different yield components on seed yield per plant. The study suggested that mutagenic treatment of F<sub>1</sub> seeds helped to break undesirable linkages. Singh and Paroda [5] found that irradiation and hybridization appeared to increase genetic variance, *gca* and *sca* than hybridization alone.

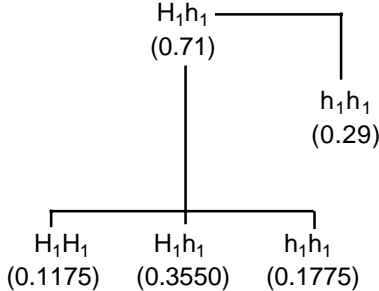
**Wilt reaction:** The segregating population of M<sub>2</sub>

of Bheema, F<sub>7</sub> (Bheema x ICCV-10) and F<sub>2</sub>M<sub>1</sub> (Bheema x ICCV-10) were evaluated in the wilt sick plots of ICRISAT. The percentage of wilted plants in F<sub>2</sub> was the highest (77.67) followed by that of M<sub>2</sub> (70.79). However, F<sub>2</sub>M<sub>1</sub> of Bheema x ICCV-10 showed much lesser value of wilt percentage (55.00). The results are on expected line indicating the advantage with F<sub>2</sub>M<sub>1</sub> compared to F<sub>2</sub> in having a highest percentage of plants which were wilt free (59.52%) compared to that of F<sub>2</sub> (22.3%) and M<sub>2</sub> (29.2%). M<sub>2</sub> showed slight advantage over F<sub>2</sub>. Changes in the frequency of resistant types in mutated population will explain how the F<sub>2</sub>M<sub>1</sub> is advantageous over both F<sub>2</sub> and M<sub>2</sub> (Table 3). When a genotype with dominant loci (Bheema) is crossed with a genotype with recessive loci (ICCV-10), the F<sub>1</sub> will be heterozygous. On generation advancement from F<sub>1</sub> to F<sub>2</sub>, the F<sub>2</sub> population will have the normal segregating pattern in the ratio of 1:2:1, where 25 % of population constitutes resistant genotype. But in mutated F<sub>2</sub> population (F<sub>2</sub>M<sub>1</sub>) the frequency is not in normal ratio of 1:2:1, instead the recessive types which are known to be resistant are increased at the cost of heterozygous types. As shown in Table 2, the M<sub>2</sub> of Bheema showed around 29% of resistant genotypes against 0% of resistant types in susceptible genotype Bheema suggesting that the rate at which mutation resulted in resistant homozygous types is 29%. Thus, the F<sub>2</sub>M<sub>1</sub> showed around 59% of resistant types against 25 % of resistant types in F<sub>2</sub> is obviously the result of combination of hybridization followed by mutation and this has additional advantage over other two populations (Table 2 and 3). On the basis of observations, F<sub>2</sub>M<sub>1</sub> with 29% mutation frequency (as that of M<sub>2</sub>) should yield around 47% of resistant types which also includes the source of recombination and segregation in F<sub>2</sub>. But the realized resistant types were 59% (Table 2 and 3) which was much higher than expected 47% resistant types. The difference of 59-47% would therefore be suspected to have aroused from higher chances of converting heterozygous loci (H<sub>1</sub>h<sub>1</sub> in F<sub>1</sub>) into resistant types than that of converting homozygous loci (h<sub>1</sub>h<sub>1</sub> in Bheema) into resistant types.

**Table 2.** Wilting (%) in breeding material recorded at ICRISAT, Hyderabad

Breeding population	No. of plants wilted	No. of healthy plants	Total no. of plants	Wilt reaction(%)	
				Resistant	Susceptible
M <sub>2</sub> of Bheema	269	111	380	29.21	70.79
F <sub>2</sub> of Bheemax ICC V- 10	445	113	573	22.33	77.67
F <sub>2</sub> M <sub>1</sub> of Bheema x 1CCV 10	170	250	420	59.52	40.48

**Table 3.** Expected segregating pattern in mutated population ( $M_1$  and  $F_2M_1$ ) based on mutation frequency in  $M_1$  against non mutated ( $F_1$ ) population in chickpea

Population	Segregating pattern	Segregation ratio Resistant : Susceptible
$P_1$	$H_1H_1 : 0 : 0$	0 : 1
$P_2$	$0 : 0 : h_1h_1$	1 : 0
$F_1$	$0 : H_1h_1 : 0$	0 : 1
$F_2$	$H_1H_1 : H_1h_1 : h_1h_1$	0.75 : 0.25
$M_2$	$H_1H_1 : H_1h_1 : h_1h_1 / H_1H_1 : h_1h_1$	0.71 : 0.29
$F_2M_1$		0.53 : 0.47

As expected, on overall basis considering mean, extent of variation and frequency of resistant types  $F_2M_1$  was found to be a better population followed by  $F_2$  and  $M_2$ . The hypothesis that combined effect of irradiation and hybridization is better in generating broader spectrum of variability has been realized.  $F_2M_1$  populations thus provide better scope for effecting selection for improving productivity. There are reports as indicated in literature [6-8] that this technique has helped in releasing more variability than realized either by hybridization or by induced mutation alone. Therefore it is concluded that integrated approach of induced mutations and hybridization has greater impact on releasing broad spectrum of variability for yield and yield related traits and also to transfer the resistance to fusarium wilt in chickpea.

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