

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 F₂ seeds of cross between Bheema and ICCV-10 were subjected to gamma irradiation. Simultaneously, F₂ was also advanced to F₃ generation. Besides, Bheema was also subjected to irradiation to look to the possibility of developing high yielding and disease resistant mutants. The F₂M₁ population proved to be highly potential in recovering higher frequency of high yielding and wilt resistant types than selfed (F₃) and mutant (M₂) 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 F₁ seeds were harvested and were grown for generation advancement during *kharif* 2008 using off-season nursery. About 250 F₂ seeds harvested from F₁ plants were subjected to γ irradiation @ 60 kr during 2008 and remnant F₂ seeds were kept under proper storage conditions. Similarly, M₁ of Bheema obtained by subjecting 250 seeds of Bheema to γ irradiation (during *rabi* 2008-09) following the same dose was advanced to get M₂ of Bheema. And also un-irradiated F₁ 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 ICRI SAT 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 γ 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

M₂ 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 F₂M₁ showed the highest CV which is closely followed by that of F₂ in respect of seed yield and pods per plant. The CV of M₂ 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, F₂M₁ was a better population than either F₂ or M₂.

Range: In respect of seed yield, the range of variation of F₂M₁ and F₂ was comparable and high compared to that of M₂. However, for number of pods per plant the range of expression of F₂M₁ was much higher compared to F₂ and that of M₂ was much less. But the range of expression for seed weight of F₂ was highest followed by that of F₂M₁ and M₂ while, upper range of expression for seed weight was highest in case of M₂. On the basis of range of expression also F₂M₁ again proved to be a better population than F₂ and M₂ 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 F₁ 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 M₂

of Bheema, F₇ (Bheema x ICCV-10) and F₂M₁ (Bheema x ICCV-10) were evaluated in the wilt sick plots of ICRISAT. The percentage of wilted plants in F₂ was the highest (77.67) followed by that of M₂ (70.79). However, F₂M₁ of Bheema x ICCV-10 showed much lesser value of wilt percentage (55.00). The results are on expected line indicating the advantage with F₂M₁ compared to F₂ in having a highest percentage of plants which were wilt free (59.52%) compared to that of F₂ (22.3%) and M₂ (29.2%). M₂ showed slight advantage over F₂. Changes in the frequency of resistant types in mutated population will explain how the F₂M₁ is advantageous over both F₂ and M₂ (Table 3). When a genotype with dominant loci (Bheema) is crossed with a genotype with recessive loci (ICCV-10), the F₁ will be heterozygous. On generation advancement from F₁ to F₂, the F₂ population will have the normal segregating pattern in the ratio of 1:2:1, where 25 % of population constitutes resistant genotype. But in mutated F₂ population (F₂M₁) 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 M₂ 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 F₂M₁ showed around 59% of resistant types against 25 % of resistant types in F₂ 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, F₂M₁ with 29% mutation frequency (as that of M₂) should yield around 47% of resistant types which also includes the source of recombination and segregation in F₂. 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 (H₁h₁ in F₁) into resistant types than that of converting homozygous loci (h₁h₁ 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 ₂ of Bheema	269	111	380	29.21	70.79
F ₂ of Bheemax ICC V- 10	445	113	573	22.33	77.67
F ₂ M ₁ 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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