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Genetics and Characterization of an Open Flower Mutant in Chickpea

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The chickpea (Cicer arietinum L.) is a self-pollinated grain legume with cleistogamous flowers. A spontaneous openflower mutant, designated OFM-3, was identified in which reproductive organs were not enclosed by the keel petals and thus remained exposed. All 10 stamens in this mutant were free, whereas these are in diadelphous (9 fused + 1 free) condition in normal chickpea flowers. A large number of pods (73%) remained unfilled (empty) in OFM-3, though its pollen fertility was as high as the standard cultivars. The open-flower trait was found to be recessive and controlled by a single gene. OFM-3 was crossed with earlier reported open-flower mutants, ICC 16341 and ICC 16129, to establish trait relationships of genes controlling open flower traits in these mutants. It was found that each of these mutants has a unique gene for open flower trait. The genes controlling open flower trait in ICC 16341, ICC 16129, and OFM-3 were designated ofl-1, ofl-2, and ofl-3, respectively. Breeding lines with open flower trait and higher percentage of filled pods have been developed from the progenies of the crosses of OFM-3 with normal-flowered lines. The open flower trait offers opportunity for exploring hybrid technology in the chickpea.

Key words: allelic relationship, Cicer arietinum, hybrids, inheritance, pod filling

Chickpea (*Cicer arietinum* L.) is a self-pollinated grain legume with cleistogamous flowers. Spontaneous and induced mutations and hybridization are the main sources of variation for chickpea breeding. Mutation breeding has been successfully used to develop 15 improved cultivars (Salimath et al. 2007). Many new traits, like triple flower per axis (Singh and Chaturvedi 1998) and multiflower per axis (Gaur and Gour 2002), were identified in spontaneous mutants that have a potential for improving seed yields by increasing number of pods per node. Two brachytic mutants, one originated spontaneously (Dahiya et al. 1984) and other induced through mutagenesis (Gaur et al. 2008), with short internodes and compact growth habit have been used in ideotype breeding and promising progenies with compact growth habit and which can be grown at high plant density have been obtained (Lather 2000; Gaur et al. 2008). Similarly, spontaneous (Knights 1993) and induced (Gaur and Gour 1999) stem fasciation mutants were established, in which pods accumulate at top of the stem and form pod clusters, and such plant type can facilitate mechanical harvest (Srinivasan et al. 2008).

The available reports on outcrossing in chickpea indicate that chickpea is a highly self-pollinated crop. The outcrossing is reported to be in the range of 0.0–1.9% (Gowda 1981; Malhotra and Singh 1986; Tayyar et al. 1995; Toker et al. 2006). Though cleistogamy in chickpea is a useful trait for maintaining genetic purity of cultivars, it poses challenges in development of hybrids. In normal chickpea flowers, the reproductive organs (stamens and gynecium) remained enclosed by the keel petals preventing cross-pollination. Flowers with all petals open (open flower) that expose male and female reproductive organs would facilitate cross-pollination. Two such open flower mutants have been reported earlier in desi chickpea. Dahiya et al. (1984) isolated an open flower mutant from the breeding line HMS 6-1 at Haryana Agricultural University, Hisar, North India (29°11"N, 75°39"E), and this mutant was later registered in the name of ICC 16341 in International Crops Research Institute for the Semi-Arid Tropics (ICRISAT)'s germplasm collection. The second mutant line ICC 16129 was developed by Pundir and Reddy (1998) at ICRISAT, Patancheru, India (17°27"N, 78°28" E) from a spontaneous mutant of the land race ICC 16127 originated from Mimbu, Magwe, Myanmar (20°80"N, 93°45"N). Inheritance of open flower trait was monogenic in ICC 16129 (Pundir and Reddy 1998) and not known in ICC 16341.

This study reports a new open flower mutant identified in a chickpea breeding line ICCV 04502 at ICRISAT, Patancheru. Morphological features, inheritance, and yield characteristics of the mutant have been described, and its utility in breeding programs has been discussed. Furthermore, the allelic relationships of present and previously reported open flower mutants have been described.

Materials and Methods

The open flower mutant (OFM-3) identified in this study was crossed with one Indian popular cultivar JG 11 (single flower per axis) and a super-early breeding line ICCV 96030 (double or twin flowers per axis) to investigate the inheritance of open flower trait and to identify segregants with desirable plant types and yield characteristics. Experiments were conducted at ICRISAT, Patancheru, and initial crosses were made in offseason in glass house during March 2006. About half of the crossed seeds were planted in field during postrainy season (October-February) 2006/2007 and allowed self-pollination for generation advancement. Parents, F1 (from the saved crossed seed), and F₂ populations were grown together in field during postrainy season 2007/2008 and F2-derived F3 progenies in 2008/2009. The crop was raised under rainfed conditions on vertisol without any supplemental irrigation. The row-to-row and plant-to-plant spacing was kept at 60 and 20 cm, respectively. The package of practices suggested by Gaur et al. (2010) was followed to grow a healthy crop and control insect pests. Observations were recorded on 366 F2 plants in OFM-3 \times ICCV 96030 and 428 F₂ plants in OFM-3 \times JG 11. OFM-3 was used as female parent in both the crosses. F₃ progenies were grown from about 100 F₂ plants in each cross. Fifteen nonsegregating F₃ progenies of each normal- and open-flowered plant types were taken in each cross for comparing yield contributing parameters between the 2 flower types.

For studying allelic relationships of genes controlling open flower trait, OFM-3 and previously reported open flower mutants ICC 16341 and ICC 16129 were crossed in all possible combinations excluding reciprocals. F_{1s} were grown under glass house in 2008, and F_{2s} were grown in field during postrainy season 2009/2010.

Inheritance of qualitative traits was studied using a computer program LINKAGE-1 (Suiter et al. 1983), which tests goodness-of-fit to expected ratios by chi-square analysis and detects the presence of linkage using a contingency chi-square test. GENSTAT (version 10.3) was used to test significance of the differences between open flower and normal-flowered segregants for various traits in F_3 progenies.

The area of standard petals was measured on 4 random plants from each genotype and by taking 5 mature flowers from each plant. A graphical method was used for measuring surface area of standard petals. The petal was placed on a graph paper with 1×1 mm grid cells. The outline of the petal was traced on the graph paper with a fine pencil. The grid cells where the petal covered 50% or more of the area of an individual grid cell were counted to calculate the surface area of the petal.

Pollen grains for viability test were taken from 4- to 5day-old flower buds. Pollen viability test was performed with acetocarmine dye where pollen stained with pink color was considered as viable. Pollen observations were taken on 10 different locations under compound microscope. Furthermore, pollen germination was tested on cellophane placed on growth media containing CaCl₂·2H₂O (300 mg l⁻¹), H₃BO₃ (100 mg l⁻¹), and sucrose 15% (Brewbaker and Kwack 1963; Alexander and Ganeshan 1989) and incubated for 8 h in darkness at 20–25 °C. Cellophane was lifted and aniline blue stain was added before observation under a microscope (Carl Zeiss, Germany). Pollen was considered to have germinated when tube length was more than the diameter of the pollen grain.

Results and Discussion

Origin and Morphological Features of Open Flower Mutant

A spontaneous open flower mutant was observed during postrainy season 2004 at ICRISAT—Patancheru in a breeding line ICCV 04502 developed from cross C 235 \times ILC 3279. This mutant bred true in the succeeding generations. The fixed line developed from this mutant (OFM-3) is being maintained by ICRISAT's chickpea breeding program.

The morphological features of OFM-3 in comparison with other open flower mutants are presented in Table 1. OFM-3 has light pink flowers. Leaves have normal green color and are imparipinnate type with average leaflet number 14 (data not shown). The 3 terminal leaflets are very close and form a cone-shaped cluster when the leaf is young. Main stem is weak, which gives mostly 2 primary branches and grows horizontally over the soil. Secondary branches appear laterally on the primary branches and grow vertically bearing more than 90% of the total flowers/pods.

Flower structure of OFM-3 is quite unique compared with normal cleistogamous (closed) flower and also different from previously reported open flower mutants ICC 16341 and ICC 16129 (Table 1, Figure 1). The corolla in normal chickpea flower is comprised of standard petal, 2 wing petals, and 2 keel petals. Keel petals enclose male and female organs throughout their development, which enforces selfpollination. In OFM-3, all the 5 petals are free as observed in ICC 16341 and ICC 16129. However, the standard petal in OFM-3 is slightly larger than that in ICC 16341 and more than double in size than in ICC 16129. As the age of flower increases, standard and wing petals expand and wide open, and the keel petals expose the anthers and stigma on dorsal side similar to ICC 16341. On contrary, keel petals in ICC 16129 open through ventral side of the flower.

Diadelphous [(9) + 1] condition of stamens is a characteristic feature of the family leguminaceae, that is, 9 stamens with their filaments fused forming an androecial sheath and the 10th stamen completely free. In contrast to this, in OFM-3, all the filaments are free throughout the length (Figure 1c). This is similar to ICC 16341 but different than ICC 16129, which has diadelphous condition (Figure 1a). In pigeonpea, open flower segregants having all 10 free stamens observed (Cherian et al. 2006). Ovary is also large in OFM-3 compared with normal flowers and flowers of ICC 16129 (Figure 1b).

Pod Filling/Seed Setting and Pollen Fertility of Open Flower Mutant

A large number of pods did not develop seeds in OFM-3 giving a high percentage (73%) of empty pods (Figure 1d).

Features	OFM-3	ICC 16341	ICC 16129		
Origin of mutation	Spontaneous	Spontaneous	Spontaneous		
Stem color	Light green	Normal green	Purplish green		
Flower color	Light pink	Pink	Pink		
Petals position	Rotate	Rotate	Fixed		
Area of standard petal (mm ²)	95.2 ± 2.74	72.2 ± 2.39	37.3 ± 0.84		
Filaments	Free	Free	(9) + 1		
Size of androecium	More than normal flower	More than normal flower	Less than normal flower		
Days to flowering	42.0 ± 0.47	46.0 ± 1.22	52.0 ± 0.28		
Plant height (cm)	42.5 ± 2.17	38.2 ± 2.32	62.2 ± 4.34		
Plant width (cm)	54.5 ± 5.15	32.5 ± 4.34	42.5 ± 3.91		
Growth habit	Spreading.(weak stem)	Semispreading (weak stem)	Semierrect		
Days to maturity	115.0 ± 0.71	107.0 ± 3.11	102.3 ± 2.45		
Number of filled pod/plant	21.0 ± 5.16	65.0 ± 7.71	168.8 ± 15.32		
Number of empty pod/plant	57.2 ± 6.10	41.3 ± 4.32	3.2 ± 0.64		
Number of seed/plant	28.8 ± 5.92	81.6 ± 10.31	213.5 ± 20.21		
Yield/plant (g)	7.7 ± 1.21	8.3 ± 1.11	15.8 ± 1.11		
100-seed weight (g)	22.4 ± 1.03	10.1 ± 0.32	7.6 ± 0.22		
% of filled pods	26.9 ± 6.10	60.7 ± 1.51	98.2 ± 0.32		
Seed color	Dark brown	Brown	Yellowish brown		

Table 1 Characteristics of newly identified open flower mutant (OFM-3) and 2 previously reported open-flowered mutants(ICC 16341 and ICC 16129)

Thus, percentage of filled pods was very low (27%) in OFM as compared with earlier reported open flower mutants ICC 16341 and ICC 16129, which produced 61%



Figure 1. (a) Flower morphology of OFM-3, ICC 16129 and their F_1 , (b) variation in size of gynecium, (c) diadelphous and monoadelphous condition of stamens in normal flower (JG 11) and OFM-3, and (d) a branch of OFM-3 with large number of empty pods.

and 98% filled pods, respectively (Table 1). Weather parameters were recorded during the crop growth period to see the effect of environment on pod filling. Except for average rainfall (44, 12, and 13 mm), there was not much difference in maximum (29, 30, and 30 °C) and minimum (14, 15, and 14 °C) temperatures and solar radiation (17, 16, and 17 mJ/m²) during crop growth period over 3 years 2005/2006, 2006/2007, and 2007/2008. The OFM-3 produced large number of empty pods in all these years, suggesting that low seed set was not due to environmental factors. The physiological processes affecting embryo development and poor seed set in OFM-3 need further investigations.

OFM-3 had larger seeds (22.1 g/100-seed) as compared with ICC 16341 (10.1 g/100-seed) and ICC 16129 (7.6 g/ 100-seed). As yield per plant is determined by number of seeds per plant and seed size, the yield of OFM-3 was similar to ICC 16341 due to large seed size. ICC 16129 gave the highest yield because of large number of filled pods per plant (Table 1).

Sexual reproduction in angiosperms often depends on the normal development of all floral parts. Abnormal development of one floral structure can disrupt development of subsequent parts and may cause sterility (Johns et al. 1981; Johns and Palmer 1982). A soybean line that lacked standard petal, lateral wings, and keel petals was found to be male sterile (Palmer et al. 2004). As OFM-3 had a high percentage of empty pods, we suspected that it might have high pollen sterility. However, pollen viability test using acetocarmine dve showed that 98% of pollen grains were fertile in OFM-3. Similarly, pollen germination test with sucrose solution revealed 96% pollen germination. This indicates that pollen fertility did not affect pod filling/seed setting in OFM-3. Similarly, Pundir and Reddy (1998) also reported that pollen viability was not a problem in ICC 16129.

Table 2 Inheritance of open flower trait in chickp
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Cross	Fi		F ₂		F ₃		Batio	~)	Р
	Normal		Open	Normal	Segregating	Open	Racio	<u>λ</u> 2	,
OFM-3 × ICCV 96030	Normal	267	99	_			3:1	0.19	0.35
				17	71	23			
$OFM-3 \times JG 11$	Normal	317	111		_		3:1	0.21	0.35
-				27	51	23			
OFM-3 × ICC 16129	Normal	132	39 (OFM-3 type) 44 (ICC 16129 type) 12 (Deformed plants)	_	_		9:3:3:1	0.85	0.16
OFM-3 × ICC 16341 ICC 16129 × ICC 16341	Normal Normal	_							

Inheritance and Allelic Relationships of Genes Controlling Open flower Trait

Inheritance of open flower trait of OFM-3 was studied in F2 of 2 crosses, OFM-3 × JG 11 and OFM-3 × ICCV 96030 (Table 2). The F1 plants produced normal cleistogamous (closed) flowers in both the crosses indicating dominance of closed flower over open flower. The F2 in each of these crosses segregated in a 3 (close flower):1 (open flower) ratio, suggesting that the open flower trait is controlled by a single gene. This was further confirmed in F3 progenies where 25% progenies bred true for close flower, 25% for open flower, and 50% showed segregation for close and open flowers. The open flower trait in ICC 16129 reported to be controlled by a single gene (Pundir and Reddy 1998), and its inheritance was not studied in ICC 16341. Joint segregation of open flower trait and double flower trait was studied in the cross OFM-3 (single flower per node) \times ICCV 96030 (2 flowers per node). The single-flowered and the doubledflowered plants gave a good fit to a 3 (283 plants):1 (83 plants) ratio with $\chi 2$ value 1.05 (P = 0.31). These results are in agreement with previous studies on genetics of double flower trait in chickpea (Khan and Akhtar 1934; Singh and van Rheenen 1994; Kumar et al. 2000; Gaur and Gour 2002). Joint segregation analysis showed that in normalflowered plants, 194 were single-flowered and 73 were double-flowered, whereas in open-flowered plants, 89 were single-flowered and 10 were double-flowered. These results deviated from the normal digenic segregation ratio of 9:3:3:1 for flower type and number of flowers per axis and showed a weak linkage (r = 0.33) between the 2 traits. In a previous study with ICC 16129 genotype, a linkage (r = 0.34) was detected between flower type and leaf size (Pundir and Reddy 1998). Each F₂ population was thoroughly examined for different plant types (semispreading and spreading), flower types (closed and open), and their recombinants. Interestingly, there was no single segregant having normal plant type with open flower trait combination observed in F₂ and F₃ of both the crosses. This clearly shows a strong linkage between plant type and open flower traits. Crossing with different genotypes and raising large segregating population may help in recovering recombinants with normal plant type and open flower trait if this linkage is not due to pleiotropy.

Allelic relationship of genes controlling open flower trait in OFM-3 and the earlier reported open flower mutants ICC 16341 and ICC 16129 was studied by crossing these mutants in all possible combinations excluding reciprocals. F1 plants from all crosses (open flower × open flower) produced normal closed flowers (Table 2). This indicates that open flower trait in these mutants is controlled by different (nonallelic) genes. The genes controlling open flower trait in these mutants are designated as follows: ofl-1 in ICC 16341 (first report on open flower), off-2 in ICC 16129, and off-3 in the present mutant OFM-3. The F2 segregation could be studied only in one cross OFM-3 × ICC 16129 because the plants died in other 2 crosses due to wilt. The F1 plants in OFM-3 \times ICC 16129 produced normal flowers because of dominance at both the loci (Off-2_Off-3_). The F2 segregated in a 9 (normal flower): 3 (ICC 16129 type open flower):3 (OFM-3 type open flower):1 (deformed flower) ratio. Dominance at both the loci produced normal flowers (Off-2_Off-3_), the dominance at only Off-3 locus produced ICC 16129 type of open flowers (off-2off-2 Off-3_), the dominance at only Off-2 locus produced OFM-3 type of open flowers (Off-2_ off-3off-3), and the double recessive (ofl-2ofl-2 ofl-3ofl-3) produced sterile deformed flowers.

Similar to this report, there are earlier reports where the same trait was reported to be expressed by mutations at different loci (nonallelic genes). For example, the spontaneous and the induced mutant genes for stem fasciation (Srinivasan et al. 2008) as well as for brachytic growth habit (Gaur et al. 2008) were found to be nonallelic. Identification of different genes for the same trait (open flower trait in this case) provides options for deploying a different gene for that trait when one gene is closely linked to an undesired gene.

Influence of Open Flower Trait on Yield and Related Traits

The open flower F_2 plants, on average, had 46.4% (range 0– 85) filled pods in OFM-3 × ICCV 96030 and 61.7% (range 0–90) in OFM-3 × JG 11. The open flower F_3 progenies gave 47.3% filled pods in OFM-3 × ICCV 96030 and 82.1% in OFM-3 × JG 11. Large variability for pod-filling percentage in open flower segregants provided an opportunity of selecting open flower plants with high percentage of pod filling. This also showed that reduced percentage of

Table 3	Comparison of normal and open flower	segregants for various	characteristics in F2 p	population and F ₃ pro	ogenies of OFM-3 \times
ICCV 960	30 and OFM-3 \times JG 11 crosses.				

	F2					F ₃		
	Normal flower		Open flower			Normal flower		
Character	$\text{Mean} \pm \text{SE}$	Range	$Mean \pm SE$	Range	t _{prob}	Mean \pm SE	Mean \pm SE	
	$OFM-3 \times ICO$	CV 96030						
Days to flowering	47.9 ± 0.53	32-67	34.8 ± 0.45	28-50	< 0.0001	_		
Days to maturity	103.4 ± 0.47	82-125	100.7 ± 0.99	80-112	0.0151	_		
Plant width (cm)	43.7 ± 1.13	15-100	34.8 ± 1.72	9-90	< 0.0001	—		
Plant height (cm)	45.7 ± 0.61	25-97	38.4 ± 1.03	16-71	< 0.0001	_		
No of primary branches/plant	2.6 ± 0.06	1-6	1.84 ± 0.07	1–3	< 0.0001	—		
No of secondary branches/plant	4.5 ± 0.13	1-15	2.3 ± 0.20	0–6	< 0.0001	—		
No of filled pods/plant	104.8 ± 4.68	6-410	10.5 ± 0.99	0–73	< 0.0001	53.2 ± 2.02	24.5 ± 3.0	
No of empty pods/plant	7.0 ± 0.59	0-70	15.0 ± 1.38	0-60	< 0.0001	2.4 ± 0.30	33.0 ± 6.6	
No of seeds/plant	116.3 ± 5.15	6-473	13.2 ± 1.12	0–79	< 0.0001	62.9 ± 2.75	32.5 ± 3.7	
Yield/plant (g)	27.24 ± 1.21	2.0116	2.9 ± 1.88	0–21	< 0.0001	14.9 ± 0.70	6.3 ± 0.8	
100-seed weight (g)	24.1 ± 0.22	13-32	22.6 ± 0.52	4-33	0.009	23.8 ± 0.55	19.3 ± 0.9	
% Filled pods	92.2 ± 0.67	13-100	46.4 ± 2.3	0-85	< 0.0001	94.9 ± 0.58	47.3 ± 2.4	
	$OFM-3 \times JG$	11						
Days to flowering	48.8 ± 0.36	34-65	41.3 ± 0.37	35–54	< 0.0001	—	—	
Days to maturity	103.9 ± 0.37	85-115	105.9 ± 0.71	90-124	0.010	—	—	
Plant width (cm)	34.5 ± 0.76	10-85	27.7 ± 1.23	10-61	< 0.0001	—	—	
Plant height (cm)	45.1 ± 0.41	21-62	37.9 ± 1.0	17-70	< 0.0001	—	—	
No of primary branches/plant	2.4 ± 0.07	1-10	1.7 ± 0.05	1–3	< 0.0001	—	—	
No of secondary branches/plant	5.9 ± 0.16	1-17	3.9 ± 0.19	0-8	< 0.0001	_	—	
No of filled pods/plant	128.0 ± 4.84	8-700	7.1 ± 0.68	0–33	< 0.0001	71.7 ± 3.58	33.5 ± 4.32	
No of empty pods/plant	6.2 ± 0.42	0-78	5.9 ± 0.73	0–47	0.6529	2.4 ± 0.47	7.1 ± 1.32	
No of seeds/plant	137.6 ± 5.32	8-796	9.1 ± 0.83	1-42	< 0.0001	78.4 ± 2.06	42.5 ± 5.02	
Yield/plant (g)	31.4 ± 1.19	2-171	2.0 ± 0.19	0.1–10	< 0.0001	18.4 ± 0.9	9.0 ± 0.84	
100-seed weight (g)	23.1 ± 0.20	8.8-40	22.7 ± 0.75	5–45	0.614	23.8 ± 0.88	21.9 ± 0.7	
% Filled pods	94.2 ± 0.42	44.3–100	61.7 ± 2.98	0–90	< 0.0001	96.6 ± 0.46	82.1 ± 1.75	

SE, standard error.

filled pods in OFM-3 could be due to other independently inherited gene(s) that affect embryo development. Open flower progenies with high percentage (>80%) of filled pods have been developed by selection in F_{3} .

Yield contributing traits were compared between normal- and open-flowered plants in F₂ and F₃ in 2 crosses, OFM-3 × JG 11 and OFM-3 × ICCV 96030 (Table 3). Open flower plants differed significantly from normal flower plants for all traits in F₂ of both the crosses, except for empty pod ($t_{prob} = 0.653$) and 100-seed weight ($t_{prob} =$ 0.614) in cross OFM-3 × JG 11. Normal-flowered plants produced 10–18 times more number of filled pods than open-flowered plants in both the crosses. The average seed yield was 8 and 16 times more in normal-flowered plants than open-flowered plants in crosses with ICCV 96030 and JG 11, respectively. Yield advantage of normal-flowered plants over open-flowered plants was further confirmed in F₃ (Table 3).

Prospects of Using Open Flower Mutant in Chickpea Improvement

The cleistogamous flowers of chickpea ensure high levels of self-fertilization. The outcrossing is reported to be less than 2% (Gowda 1981; Malhotra and Singh 1986; Tayyar et al.

1995; Toker et al. 2006). The open flower mutants are expected to have higher levels of outcrossing as reproductive organs are fully exposed to air and insect pollinators. A study conducted on rate of outcrossing in F₃ open flower progenies derived from 2 crosses (ICC 16341 × CA 1938, ICC 16341 × ICC 4973), which had one open flower common parent (ICC 16341) revealed 5.9% outcrossing in these progenies (Rubio et al. 2010). There is a need of assessing extent of outcrossing in other open flower mutants (ICC 16129, OFM-3). The crosses between open flower mutants may give transgressive sergeants with high degree of outcrossing.

The open flower mutants open opportunity for exploring hybrid technology for chickpea improvement. Cytoplasmic male sterility (CMS) system can be developed through interspecific hybridization and combined with open flower trait. The male sterility can facilitate studies on male developmental pathway and make it possible to exploit heterosis to obtain high-yielding hybrids (Kaul 1988). Pigeonpea is an often cross-pollinated grain legume where CMS systems have been successfully developed through interspecific hybridization, and hybrids are now under commercial cultivation (Saxena and Nadarajan 2010; Saxena et al. 2010). Male sterility has been reported in chickpea, and genetic studies indicated that the male sterility was under the control of monogenic recessive gene (Chaudhary et al. 1970; Reddy and Reddy 1996). However, it is difficult and uneconomical to exploit genetic male sterility for development of commercial hybrids in chickpea where seed multiplication ratio is low (1:10–1:20) and required seed rate for sowing is high (>60 kg per ha). It would be worth exploring development of CMS system through wide crosses and combine it with open flower trait for developing hybrid technology for chickpea.

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