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IDENTIFICATION OF GENETIC LINKAGE BETWEEN MALE STERILITY AND DWARFNESS IN SAFFLOWER

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ABSTRACT

Six spontaneous dwarf male sterile (DMS) mutants of about 30 cm mean plant height were identified in safflower accession BLY 1035. The inheritance of male sterility and dwarfness in F_1, F_2 and F_3 progenies of DMS crosses suggested that the two characters are governed by different recessive genes in homozygous state. Male sterility and dwarfness are designated by the gene symbols ms_2 and dw respectively. The genetic analysis revealed close linkage between ms_2 and dw genes. Among the crosses, DMS x HUS 282 and DMS x BLY 1035 exhibited complete linkage between the sterility and dwarfing genes. However, the crosses DMS x CTV 244 and DMS x Bhima recorded 0.72 ± 0.73% and 7.97 ± 1.98% linkage distances, respectively, between ms_2 and dw. Linkage between the genes for male sterility and dwarfness is reported for the first time, and its usefulness in hybrid seed production is discussed.

Key words: Linkage, male sterility, dwarfness, allele, inheritance.

Genetic male sterility has been reported in safflower by several workers [1–8]. But the lack of a sterility-linked marker trait at early stage of plant growth has restricted its utilization in hybrid development. Therefore, the high heterosis which exists in safflower, has remained unutilized. An attempt was made earlier [2] to detect linkage between male sterility, dwarfness and pericarp types, but no linkage was observed between the genes governing these traits. Recently, relationship has been reported between trigenic male-female sterility and oil quality in safflower [7]. Only one specific gene locus (s_1) causing sterility was linked to the gene (ol) controlling oil quality. But the linkage of the male sterility gene with the gene for oil quality does not seem to have any apparent role in hybrid seed production technology. Therefore, there was a need for marker assisted identification of male sterile line at early crop stage for the success of hybrid safflower. In this context, the present study reports a case of linkage between male sterility and dwarfing genes in safflower.

MATERIALS AND METHODS

Six dwarf plants of about 30 cm plant height appeared in the 100 cm tall safflower genotype BLY 1035 during rabi 1992–93 at the Nimbkar Agricultural Research Institute, Phaltan. The dwarf plants produce slightly spreading, woody, thick and short branches with dark green leaves. The number of capitula per plant ranged from 15-20 with a pinched opening, which restricted the emergence of florets, while the stigma and anthers protruded out as usual. The anthers were full of sterile pollen grains, which was confirmed by staining in 1:1 acetocarmine-glycerol solution. The sterility was further confirmed by absence of seed setting in the forced self- pollinated capitula and in the capitula of other genotypes obtained after crossing with the pollen of dwarf plants. However, seed setting in the dwarf plants was normal when crossed with the genotypes CTV 244, HUS 282, BLY 1035 and Bhima, which confirms female fertility. The F_1 plants of 4 such crosses were raised in 2-row plots of 5 m length during summer 1993. All the F_1 plants of the respective crosses were tall and fertile. Two plants of each cross were bagged to get selfed seed. The selfed F_1 seed was used to raise the F_2 population during rabi 1993–94. The F_2 seeds were sown in 12-row plots of 5 m length with 20 and 45 cm spacings between plants and rows, respectively. The F_2 crosses were classified on the basis of their height and fertility status. About 17–40 randomly selected fertile and tall plants in each F_2 population were selfed and threshed separately to study segregation for male sterility and dwarfness in F_3 generation. The DMS plants with big capitula, wide opening and early flowering were maintained by sibmating with tall fertile plants having similar characteristics. The F_3 progenies of the crosses were raised separately from the selfed seed of F_2 plants in 2-row plots of 5 m length during rabi 1994–95. The F_3 populations of the crosses were examined for intra- and interprogeny segregation for sterility and plant height. The phenotypic segregations for these morphological traits recorded in F_2 and F_3 generations were subjected to χ^2 test. The linkage was estimated by the maximum likelihood method [9].

The genetic male sterile lines UC-148 [4], GMS-5 (A) [6], MSN-1 and MSV-1 [8] of monogenic recessive nature as reported earlier, maintained at the center, were crossed with one of the uniform sibmated progenies of dwarf male steriles to study the allelic reaction between the sterility causing genes of DMS and others mentioned as above. The F_1 generations of these crosses were raised each in a single row of 5 m length during summer 1995 and were examined for male sterility at flowering.

RESULTS AND DISCUSSION

The fertile and tall F_1 plants in all the crosses suggest recessive nature of male sterility and dwarfness in the mutants (Table 1). In F_2 generation, the segregation was in the ratio of 3 fertile : 1 sterile and 3 tall : 1 dwarf plants, indicating that both the characters were caused by single recessive genes. This was further supported by the segregation of F_3 progenies

Table 1. Inheritance of male sterility and dwarfness in F_1 and F_2 of different crosses of safflower

Cross	Generation	Total plants	Male sterility				Dwarfness			
			No. of plants		fit to 3:1 ratio		No. of plants		fit to 3:1 ratio	
			MF	MS	χ^2	P	tall	dwarf	χ^2	P
DMS x CTV 244	F_1	14	14	—	—	—	14	—	—	—
	F_2	135	98	37	0.417	0.90–0.50	99	36	0.200	0.90–0.50
DMS x HUS 282	F_1	57	57	—	—	—	57	—	—	—
	F_2	163	128	35	1.081	0.50–0.20	128	35	1.082	0.50–0.20
DMS x BLY 1035	F_1	46	46	—	—	—	46	—	—	—
	F_2	56	45	11	0.857	0.50–0.20	45	11	0.857	0.50–0.20
DMS x Bhima	F_1	31	31	—	—	—	31	—	—	—
	F_2	207	146	61	2.204	0.20–0.10	162	45	1.174	0.50–0.20

(Table 2). The F_3 progenies raised from selfed male fertile and tall F_2 plants showed the expected ratio of 1 true breeding fertile tall : 2 segregating for both traits. The pooled analysis over the F_3 lines of the respective crosses segregating for male sterility gave a good fit to 3 fertile : 1 sterile ratio (Table 3). Similarly, in case of dwarfness, pooled data of segregating F_3 lines of all the individual crosses exhibited a good fit to a 3:1 ratio of tall and dwarf plants.

In conclusion, these results provide a strong evidence that male sterility and dwarfism in the mutant plants are both governed by a single nuclear gene each in homozygous state.

Table 2. F_3 segregation for male sterility and dwarfness in the progenies raised from random sample of tall male-fertile F_2 plants from four crosses in safflower

Cross	Total lines	Male sterility				Dwarfness			
		No. of F_3 lines		fit to 1:2 ratio		No. of F_3 lines		fit to 1:2 ratio	
		true breeding fertile	segregating	χ^2	P	true breeding tall	segregating	χ^2	P
DMS x CTV 244	40	14	26	0.050	0.90–0.50	17	23	1.515	0.50–0.20
DMS x HUS 282	29	9	20	0.068	0.90–0.50	10	19	0.017	0.90–0.50
DMS x BLY 1035	17	4	13	0.737	0.50–0.20	6	11	0.029	0.90–0.50
DMS x Bhima	40	10	30	1.249	0.50–0.20	10	30	1.249	0.50–0.20
Pooled	126	37	89	0.893	0.50–0.20	43	83	0.035	0.90–0.50

Table 3. Pooled segregation for male fertile/sterile and tall/dwarf plants in the segregating F_3 progenies of four crosses in safflower

Cross	Male sterility						Dwarfness					
	No. of F_3 progenies	No. of plants			fit to 3:1 ratio		No. of F_3 progenies	No. of plants			fit to 3:1 ratio	
		total	MF	MS	χ^2	P		total	tall	dwarf	χ^2	P
DMS x CTV 244	26	732	558	174	0.590	0.50–0.20	23	663	509	154	1.110	0.50–0.20
DMS x HUS 282	20	586	452	134	1.422	0.30–0.20	19	536	404	132	0.039	0.90–0.50
DMS x BLY 1035	13	402	310	92	0.958	0.50–0.20	11	371	289	82	1.660	0.20–0.10
DMS x Bhima	30	1459	1077	382	1.087	0.50–0.20	30	1459	1080	379	0.742	0.50–0.20
Pooled	89	3179	2397	782	0.273	0.90–0.50	83	3029	2282	747	0.185	0.90-0.50

Male sterility in safflower has been reported as a monogenic recessive trait [1, 2, 4, 6, 8] for which the genes were designated as ms [4] and ms_1 [8]. The allelic relationship between the DMS and other male sterile mutants was studied in their respective crosses (Table 4). All the crosses with dwarf male sterile line produced only fertile and tall plants in their F_1 generations, indicating that the male sterility gene in DMS is different from the ms gene in UC-148, MSV-1 and MSN-1 and ms_1 gene in GMS-5 [A]. Therefore, the gene for male sterility in the present study is designated as ms_2. Dwarfism is also reported as a monogenic recessive trait [2], however, no gene symbol is assigned for this trait. Hence, for dwarfism in the DMS mutants reported in the present study, gene symbol dw is proposed.

Table 4. Frequency of fertile and sterile plants in DMS crosses with different male sterile mutants of safflower

Cross	Male fertile	Male sterile
DMS x UC-148	11	—
DMS x GMS-5 (A)	18	—
DMS x MSN-1	22	—
DMS x MSV-1	15	—

The joint phenotypic segregation of male sterility and dwarfness in four crosses showed highly significant χ^2 values, indicating linkage between the genes governing the two traits in coupling phase (Table 5). The maximum likelihood estimate of the map distance between ms_2 and dw was 7.97 ± 1.98% for the cross DMS x Bhima, followed by 0.72 ± 0.73% in DMS x CTV 244. However, the crosses DMS x HUS 282 and DMS x BLY 1035 showed complete linkage between the two genes. The linkage of male sterility with dwarfness in safflower is reported for the first time.

The detection of linkage between the genes for male sterility and dwarfness has made identification and roguing of male fertile plants in the dwarf male sterile lines possible about

Table 5. Joint segregation of male sterility and dwarfness and their recombination frequency in different crosses of safflower

Cross	O/E	Total F_2 plants	F_2 frequency				χ^2 (9:3:3:1)	P	Recombination fraction (%)	SE %
			AB	Ab	aB	ab				
DMS x CTV 244	O	135	98	0	1	36				
	E		75.96	25.32	25.32	8.44	145.68	>0.001	0.72	0.732
DMS x HUS 282	O	163	128	0	0	35				
	E		91.62	30.54	30.54	10.18	136.039	>0.001	0.00	0.00
DMS x BLY 1035	O	56	45	0	0	11				
	E		31.5	10.5	10.5	3.5	42.857	>0.001	0.00	0.00
DMS x Bhima	O	207	146	0	16	45				
	E		116.46	38.82	38.82	12.94	139.158	>0.001	7.97	1.977

Phenotypes: AB — both genes dominant; Ab — first gene dominant, second gene recessive; aB — first gene recessive, second gene dominant; and ab — both genes recessive.

40 days after sowing. In other GMS sources this was only possible after flowering. Therefore, the DMS lines have an added advantage over the lines of other GMS sources in the context of hybrid seed production. They may have a significant role in the commercialization of safflower hybrids.

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