



Research Note

Morphophysiological expression in *cms* analogues of sunflower (*Helianthus annuus* L.) Under water stress environment

Vikrant Tyagi, S. K. Dhillon and B. S. Gill

Deptt. of Plant Breeding and Genetics, PAU, Ludhiana, India.

E-mail: vikranttyagi97@gmail.com

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Abstract

The objective of this study was to evaluate the effect of water stress on morphological, seed yield and quality traits of sunflower *cms* analogues. The material comprised nine *cms* analogues having different cytoplasmic background namely CMS-XA (Unknown), E002-91, PKUZ-A, ARG-2A (*H. argophyllus*), ARG-3A (*H. argophyllus*), ARG-6A (*H. argophyllus*), DV-10A (*H. debilis ssp vestitus*), PHIR-27A (*H. praecox ssp hirtus*) and PRUN-29A (*H. praecox ssp runyonii*) with a common maintainer line (NC-41B). The material was grown during spring season 2011 and 2012 in randomized block design with three replications in a plot size of 4.5 m. × 0.6m. To create water stress environment the irrigation was stopped after the anthesis was complete. The data were recorded on different morphological, seed yield and quality traits. The analysis of variance revealed highly significant differences for all the traits over the years. The *cms* analogues CMS-XA (37.48g), E002-91(32.07g), ARG-3A (32.17g), PHIR-27A (34.03g) and PRUN-29A (30.42g) were observed to be significantly higher yielder than that of NC-41B (23.53g). This suggested that these sources might be exploited to develop water use efficient *cms* lines suitable for developing hybrids for growing under water stress conditions.

Keywords

Sunflower, wild source, alloplasmic *cms* lines and water stress

Sunflower is considered to be an important oilseed crop due to its rich oil composition and high nutrition quality. Sunflower grain yield, an important economic and complex trait, its inheritance depends upon a different traits which are often polygenic in nature and highly affected by environmental situations (Nadarajan and Gunasekaran, 2005). The knowledge of genetic variation presents within populations are very helpful for the efficient use of genetic resources in breeding program (Safavi et al, 2010). Recently, several *cms* backgrounds have been developed by interspecific and intraspecific crosses which resulted in more than 70 *cms* sources (Series, 2002). Since these *cms* sources were recognized, several experiments to estimate the influence of the cytoplasmic impact on important agronomic, yield and quality traits have been developed before their introgression into commercial breeding programmes. It has been very useful to broaden the genetic and cytoplasmic variability of cultivated sunflower. A remarkable success was obtained for some traits as in the case of the seed weight increase developed by Domingen, which has made possible the development of high seed yield cultivars. Water deficiency is becoming a main problem for sustainable agriculture in India. The reduced rainfall, simultaneously with high evapotranspiration is probable to subject natural and agricultural vegetation to a great risk of severe and prolonged water stress with each passing year (Ellsworth, 1999). Water stress, particularly in

sunflower at vegetative phase of the plant may result in 61% yield reduction (Iqbal, 2004). So, keeping in view above points the present study was plan to evaluation of different *cms* sources under water stress condition. At PAU a set of nine alloplasmic *cms* lines were developed from different cytoplasmic sources using NC41B as common maintainer for all these sources using backcross method. Performance of these sources for morphophysiological, yield and quality traits under normal irrigated environment has already been reported by Tyagi *et al* 2013 and 2015.

Nine interspecific crosses (F₁'s) representing different sources along with one common maintainer line NC41B were obtained from Directorate of Oilseed Research, Hyderabad. To obtain *cms* analogues all derivatives were crossed with maintainer line NC-41B followed by repeated backcrossing. Both spring (January to July) and off seasons (August to December) were exploited for attempting back crosses. The phenotypic uniformity with respect to morphological characters within these *cms* analogues was obtained in BC₇/BC₈ progenies. The obtained *cms* analogues were grown during spring season 2011 and 2012 in randomized block design with three replications in a plot size of 4.5 m. × 0.6m under water stress environment. The irrigation was stopped after the anthesis for stress environment and all the *cms* analogues were evaluated for main morphological, agronomic, physiological and quality traits. The

data were recorded for days to flower initiation, days to 50 percent flowering, days to maturity, plant height (cm), head diameter (cm), number of leaves per plant, leaf area (m²), specific leaf weight (g), leaf area index, leaf water potential (mpa), relative leaf water content (percent), photosynthetic efficiency (SPAD reading), proline content (mg/g dry weight of leaf), 100 seed weight (g), seed yield (g), biological yield (g), harvest index, oil content (percent) and fatty acid composition separately for two years 2011 and 2012 and pooled over the years. The data for seed yield, oil and quality were taken from randomly selected ten open pollinated heads of *cms analogues*. Oil content was estimated using Nuclear Magnetic Resonance (NMR) and fatty acid profiles were estimated using Gas Liquid Chromatography (GLC). The data were subjected to statistical analysis as per standard statistical protocol. The variance components and coefficients of variation were computed as per Burton (1952). The heritability in broad sense and expected genetic advance were determined by using the formula given by Johnson *et al.* (1955).

The analysis of variance revealed significant differences among these *cms analogues* for all the traits (Table 1). The pooled analysis of variance over years indicated highly significant mean squares due to *cms analogues* and years for all the traits except head diameter for which the years did not have significant effects. Highly significant *cms analogues* x year's interactions for all the traits showed differential behaviour of *cms* over the years except for relative leaf water content.

Since these *cms analogues* had same nuclear genotype, these differences could be attributed due to differences in cytoplasmic genes/factors and interaction of cytoplasmic genes and nuclear genes.

Morphological and seed yield traits: The mean performances of *cms analogues* with respect to morphological and seed yield related traits were presented in Table 2. *CMS analogues* PRUN-29A, CMS-XA and E002-91A, ARG-3A and DV-10A were recorded as late maturing than NC-41B. This indicated that the cultivated *cms* source PET-1 had shorter reproductive phase and longer vegetative phase as compared to other *cms analogues*, which, derived from different wild sources of sunflower having long duration for reproductive phase as compared to PET-1. All the *cms analogues* recorded significantly tall and bigger head diameter as compared to NC-41B. The differences among the *cms analogues* were observed to be significant. *CMS* E002-91 and ARG-3A were observed to have bigger head size among all nine alloplasmic *cms* lines. The *cms* PRUN-29A, *cms* E002-91A, *cms* ARG-3A, *cms* PKU-2A and *cms* ARG-6A recorded higher 100 seed weight as compared to NC-41B.

The *cms analogues* CMS-XA, E002-91, ARG-3A, PHIR-27A and PRUN-29A observed to be significantly higher yielding than NC-41B. This suggested that these sources might be exploited to develop water use efficient *cms* lines suitable for breeding hybrids suitable for water stress conditions. All the *cms analogues* had significantly higher biological yield than NC-41B except ARG-6A and PHIR-27A. *CMS* PHIR-27A and NC-41B observed high harvest index and PHIR-29A while E002-91 recorded lowest H. I.

Physiological traits: The data presented in table 2 reveals that all the *cms analogues* had significantly higher number of leaves per plant as compared to NC-41B. Number of leaves per plant was highest in *cms* PRUN-29A, while *cms* PKU-2A had the minimum number of leaves per plant. Sources, CMS-XA, E002-91A, ARG-3A and PRUN-29A had significant higher values for leaf area and leaf area index than NC-41B. The *cms analogues* CMS-XA, E002-91A and ARG-3A had higher specific leaf weight than NC-41B. The *cms* PKU-2A was the only wild source, which had significantly higher relative leaf water content than NC-41B. It was observed that all the *cms analogues* had significant higher photosynthetic efficiency than NC-41B. *CMS* DV-10A was unique source having significantly higher value for proline content.

Quality traits: Sunflower is categorized as low to medium drought sensitive crop. It has been observed that both quantity and distribution of water has a significant impact on oil yield in sunflower (Reddy *et al.* 2003 and Iqbal *et al.* 2005). Oil content was significantly different among the studied *cms analogues*. The highest oil content was recorded for *cms* PRUN-29A. All the *cms analogues* except two *i.e.* ARG-6A and PHIR-27A had significantly higher oil content than NC-41B. There was no significant difference among the *cms analogues* and NC-41B with respect to Palmitic acid. The *cms analogues* CMS-XA, PKU-2A, ARG-2A, ARG-3A and DV-10A significantly differed for Stearic acid from NC-41B. The *cms analogues* E002-91A, ARG-3A, DV-10A and PRUN-29A recorded significantly higher oleic acid content than NC-41B (Table 2).

Genetic components for morphophysiological and quality traits (pooled over years): Genetic advance, heritability, genotypic coefficient of variance (GCV) and phenotypic coefficient of variance (PCV) were computed (Table 2). Maximum heritability percent was recorded for quality traits *viz.*, oleic acid followed by stearic acid. Among morphological traits, head diameter recorded highest value for heritability, whereas days to

maturity showed minimum heritability followed by oil content and seed yield recorded moderate heritability. Genetic advance was observed to be maximum for biological yield and minimum for days to maturity. Biological yield recorded highest GCV and PCV while lowest was observed for days to maturity. Similar observations had been made earlier by Iqbal et al. (2009) and Tyagi et al. (2015) in conformity of these results under normal irrigated environment. These results suggests that seed yield per plant had high magnitude of broad sense heritability, which advocates the possibility of improvement of this traits through selection. Plant height exhibited highest genetic advance indicating its responsiveness to selection under water stress conditions. GCV and PCV were highest for leaf area, leaf area index, specific leaf weight and harvest index which indicated maximum amount of variability to be subjected to selection for these traits. The oil content revealed lower GCV and PCV which was an indication of limited scope for selection of this trait due to inadequate variability and implied the need to introgress desirable genes from diverse genetic resources through introduction and hybridization with germplasm. High heritability estimates associated with high genetic advance as percent mean was also earlier reported by Safavi et al. 2015.

Conclusion

The differences in performances of maintainer NC-41B with the *cms* analogues with respect to morphological, physiological and yield related traits might be attributed to the effect of different cytoplasmic sources or interactions of these cytoplasm with the nuclear genotype (nuclear genotype being same in all *cms* alloplasmic lines and maintainer NC-41B). The *cms* analogues CMS-XA, E002-91, ARG-3A, PHIR-27A and PRUN-29A were observed to be significantly higher yielder than NC-41B (conventional *cms* source) in water stress environment suitable for development of water use efficient sunflower hybrid based on different cytoplasmic backgrounds.

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Table 1: Analysis of variance for morphophysiological, yield and quality traits with respect to alloplasmic lines under water stress environment (individual and pooled over years)

Source	d.f.	Mean Squares									
		1	2	3	4	5	6	7	8	9	10
2011											
Replicates	2	3.60	8.24	7.24	107.04	4.29	3.70	0.00	0.01	0.09	7.04
CMS Sources	9	28.38**	18.82*	27.17**	1134.23**	53.96**	37.65**	0.98**	4.82**	29.76**	204.13**
Error	18	1.34	4.08	1.49	23.74	1.14	2.40	0.002	0.02	0.05	19.59
2012											
Replicates	2	1.63	6.03	1.63	73.98	0.26	2.50	0.00	0.01	0.04	15.77
CMS Sources	9	10.77**	8.97**	17.94**	1086.69**	41.63**	12.10**	0.15**	1.66**	4.68**	381.02**
Error	18	1.04	0.92	1.04	14.83	0.22	0.56	0.002	0.02	0.06	9.49
Pooled over years 2011 and 2012											
Rep. (Within years)	4	2.61	7.13	4.43	90.50	2.27	3.10	0.00	0.00	0.06	11.40
Years (Y)	1	22.84**	35.25**	976.06**	5065.56**	0.21	54.14**	1.85**	2.32**	56.47**	710.07**
CMS Sources (S)	9	18.78**	16.66**	16.49**	2025.10**	91.95**	37.14**	0.85**	5.36**	25.77**	566.53**
Interaction (Y x S)	9	20.36**	11.12**	28.62**	195.82**	3.63**	12.59**	0.28**	1.11**	8.66**	18.62
Error	36	1.19	2.50	1.26	19.28	0.68	1.47	0.00	0.01	0.05	14.54

*, ** - significant at 5 % and 1 % level respectively

1. Days to flower initiation, 2. Days to 50 percent flowering, 3. Days to maturity, 4. Plant height (cm), 5. Head diameter (cm), 6. Number of leaves per plant, 7. Leaf area (m²)/plant, 8. Specific leaf weight (g), 9. Leaf area index, 10. Relative leaf water content (percent),

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Table 1: (Contd...)

SOURCE	d.f.	Mean Squares										
		11	12	13	14	15	16	17	18	19	20	21
2011												
Replicates	2	3.83	0.01	0.06	12.03	848.73	0.10	0.92	0.44	0.02	0.37	0.59
CMS Sources	9	69.90**	2.11**	1.15**	134.61**	37811.40**	611.08**	30.01**	1.48**	12.32**	97.00**	100.83**
Error	18	1.65	0.003	0.20	4.29	568.59	22.67	1.51	0.16	0.09	0.30	0.55
2012												
Replicates	2	32.40	0.0003	0.13	1.03	931.25	6.97	0.02	0.62	0.70	0.34	0.71
CMS Sources	9	30.71**	0.001*	4.07**	164.40**	128929.40**	468.60**	5.88**	13.35**	12.65**	119.05**	176.78**
Error	18	0.05	0.0002	0.28	5.35	421.47	13.71	0.12	0.53	0.22	0.32	0.90
Pooled over 2011 and 2012												
Rep. (Within years)	4	18.11	0.00	0.09	6.53	889.98	3.53	0.46	0.52	0.36	0.35	0.64
Years (Y)	1	37.44**	1.36**	16.22**	48.05**	175644.50**	179.71**	28.01**	2.64**	0.37*	22.50**	6.29**
CMS Sources (S)	9	86.93**	1.05**	3.98**	258.82**	143973.90**	845.63**	15.15**	9.18**	24.38**	209.83**	255.03**
Interaction (Y x S)	9	13.67**	1.05**	1.23**	40.18**	22766.90**	234.03**	20.73**	5.64**	0.58**	6.21**	22.57**
Error	36	0.85	0.002	0.23	4.81	495.02	18.18	0.81	0.34	0.15	0.31	0.72

*, ** - significant at 5 % and 1 % level respectively

11. Photosynthetic efficiency (SPAD readings), 12. Proline content (mg/g dry weight of leaf), 13. 100 Seed weight (g), 14. Seed yield per plant (g), 15. Biological yield per plant (g), 16. Harvest index, 17. Oil content (percent), 18. Palmitic acid (percent), 19. Stearic acid (percent), 20. Oleic acid (percent), 21. Linoleic acid (percent)

Table 2: Mean performance and genetic parameters of alloplasmic *cms* lines for morphophysiological, yield and quality traits under water stress environment (pooled over the years)

		1	2	3	4	5	6	7	8	9	11
1	CMS-XA	65*	68	101*	88.17*	13.08*	18.42*	1.56*	3.92*	8.59*	65.53
2	E002-91A	64*	69	101*	111.25*	22.16*	15.38	1.04*	3.00*	5.73*	50.00
3	PKU-2A	66	69	98	62.42	15.48*	14.08	0.48	1.58	2.65	74.57*
4	ARG-2A	68	70	98	104.13*	13.19*	16.79	0.59	1.61	3.26	59.02
5	ARG-3A	65*	68	99*	105.67*	20.09*	18.79*	0.89*	2.13*	4.88*	61.01
6	ARG-6A	65*	67*	97	104.25*	13.48*	18.08*	0.49	1.32	2.68	65.18
7	DV-10A	67	68	100*	84.96	14.75*	17.63*	0.48	1.20	2.63	48.78
8	PHIR-27A	70*	73*	99*	64.58	14.93*	16.42	0.39	1.14	2.15	47.38
9	PRUN-29A	66	69	102*	105.21*	18.71*	23.13*	0.65*	1.39	3.59*	46.88
10	NC-41B (C)	68	70	97	73.51	8.69	15.67	0.33	0.98	1.82	66.58
CD 5%		1.27	1.19	1.19	13.14	2.80	1.78	0.27	0.67	1.48	6.95
Mean		66.42	69.0	99.10	90.41	15.46	17.44	0.69	1.83	3.80	58.49
Range	Min.	64	67	97	62.42	8.69	14.08	0.33	0.98	1.82	46.88
	Max.	70	73	102	111.25	22.16	23.13	1.56	3.92	8.59	74.57
h ² (%)		31.33	32.99	19.43	85.75	92.24	60.08	69.41	78.21	69.23	80.29
GA%		2.63	2.47	1.17	38.23	49.74	21.60	90.45	92.03	90.29	18.36
PCV		4.07	3.64	2.92	21.64	26.18	17.46	63.26	57.12	63.31	11.10
GCV		2.28	2.09	1.29	20.04	25.14	13.53	52.70	50.51	52.68	9.94

1. Days to flower initiation, 2. Days to 50 percent flowering, 3. Days to maturity, 4. Plant height (cm), 5. Head diameter (cm), 6. Number of leaves per plant, 7. Leaf area (m²)/plant, 8. Specific leaf weight (g), 9. Leaf area index, 10. Relative leaf water content (percent),

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Table 2: (Contd...)

S. N.	Genotypes	11	12	13	14	15	16	17	18	19	20	21
1	CMS-XA	37.18*	0.54	4.10	37.48*	275.33*	14.13	27.86*	6.18	7.63*	48.26	37.93
2	E002-91A	36.38*	0.72	5.67*	32.07*	477.00*	7.16	28.48*	5.90	2.78	59.59*	31.73
3	PKU-2A	33.22*	0.68	4.67*	20.80	227.20*	10.66	28.81*	3.31	6.22*	47.24	43.23
4	ARG-2A	35.86*	0.75	3.85	17.47	190.33*	9.45	29.28*	3.47	5.39*	46.68	44.46
5	ARG-3A	41.88*	1.03	5.15*	32.17*	377.50*	9.58	29.84*	4.99	6.58*	54.09*	34.34
6	ARG-6A	40.23*	1.01	4.67*	24.20	131.83	18.47	26.66	6.25	4.74	48.41	40.60
7	DV-10A	39.73*	1.18*	4.37	22.75	219.00*	10.88	29.14*	6.75	9.16*	51.46*	32.63
8	PHIR-27A	41.28*	0.58	3.59	34.03*	99.33	40.30	26.72	5.68	3.72	38.06	52.54*
9	PRUN-29A	39.55*	0.81	6.03*	30.42*	522.33*	6.17	30.88*	3.88	4.00	54.28*	37.85
10	NC-41B (C)	29.91	0.94	3.82	23.53	69.81	34.84	25.77	5.70	3.58	45.16	45.57
CD 5%		2.72	0.30	0.58	4.70	110.79	8.49	1.14	0.89	1.44	4.23	4.66
Mean		37.52	0.82	4.59	27.49	258.97	16.16	28.34	5.21	5.38	49.32	40.09
Range	Min.	29.91	0.54	3.59	17.47	69.81	6.17	25.77	3.31	2.78	38.06	31.73
	Max.	41.88	1.18	6.03	37.48	522.33	40.30	30.88	6.75	9.16	59.59	52.54
h ² (%)		39.88	57.50	77.59	82.40	68.09	26.47	47.93	94.30	95.87	89.10	60.89
GA%		29.27	26.15	42.34	109.91	120.30	4.91	31.15	74.60	24.10	31.31	12.63
PCV		35.63	22.08	26.49	64.75	85.77	9.01	31.55	38.40	12.20	17.06	10.07
GCV		22.50	16.74	23.34	58.78	70.77	4.64	21.84	37.29	11.95	16.10	7.86

11. Photosynthetic efficiency (SPAD readings), 12. Proline content (mg/g dry weight of leaf), 13. 100 Seed weight (g), 14. Seed yield per plant (g), 15. Biological yield per plant (g), 16. Harvest index, 17. Oil content (percent), 18. Palmitic acid (percent), 19. Stearic acid (percent), 20. Oleic acid (percent), 21. Linoleic acid (percent)