
CROP
PRODUCTION

Using A3, A4, and 9E CMS Types in Breeding Grain Sorghum Hybrids

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Received April 18, 2018

Abstract—The results of the evaluation of the combining ability (GCA and SCA effects; SCA dispersions) of the sorghum CMS lines in the test crosses have been reported. Inclusion of the Zheltozernyi 10 genome maternal lines differing in the type of the sterile cytoplasm (A3, A4, and 9E) into the crossing program can provide the opportunity to perform the pure genetic analysis of the cytoplasmic effects on the combining ability. Eighteen pollinators of the grain sorghum selected in the Rossorgo Russian Research and Project-Technological Institute of Sorghum and Corn were used as the paternal parents. A significant influence of the 9E cytoplasm on the GCA effects of the maternal line inflorescence length was revealed. The dominance additive genetic effects in the genetically controlled traits of the CMS lines (plant height, inflorescence and labellum parameters, and grain crop yield) were recorded. The 9E Zheltozerno 10/Magistr hybrid is recommended for the competitive variety trials, while the seven combinations are advised to be included into the three-line hybrid system.

Keywords: grain sorghum, combining ability, cytoplasmic effect, economically important traits

DOI: 10.3103/S1068367418060071

INTRODUCTION

At present, agricultural hybrid crop selection is based on cytoplasmic male sterility (CMS). Most hybrids of the field crops are industrially produced with the use of one type of sterility. However, the lack of sources for CMS may cause the secession of their cultivation in case of a weak resistance to diseases and pests. Therefore, researchers draw attention to revealing the CMS types in order to increase the genetic diversity of the F₁ hybrids and the opportunity of their application in breeding. Interspecific and interracial hybridization between the crop specimens reveals the CMS-inducing cytoplasm different in response to the tester restorers of fertility, morphological and histological structures of anthers, the pollen degeneration phase, and the mitochondrial and chloroplast genome structures. It is well known that the parent forms of high combining capacity should be included into breeding the hybrids of a high heterotic pattern. According to the literature datasets, the cytoplasmic effects on the combining ability of the CMS alloplasmic lines (different only in the cytoplasm type) are recorded in rice [8], wheat [9], switchgrass [10], and sorghum [11]. The combining capacity of the sorghum CMS lines produced with the use of A1 and A2 sterile cytoplasm was assessed. Therefore, determining the cytoplasmic effects on the CMS-line combining capacity, based on the A3, A4, and 9E cytoplasm types, is actually required.

MATERIALS AND METHODS

The F₁ hybrids (54 plants in total) and the parent varieties were grown in the experimental field of the Rossorgo Russian Research and Project-Technological Institute of Sorghum and Corn in 2015–2017. The plot area was 7.7 m². The plots were randomized. The repetition of the experiment was threefold. The plant density was determined manually (100 000 plants per hectare). The assessment of the F₁ hybrid selected valuable traits and the crop yield records were performed according to the common methods [12].

The A3 Zheltozerno 10, A4 Zheltozerno 10, and 9E Zheltozerno 10 maternal lines obtained from the repeated backcrossing of Zheltozerno 10 (Zh-10) with the A3 T×398, A4 T×398, and 9E T×398 CMS lines carrying the cytoplasm of the A3 (IS1112C), A4 (IS7920C), and 9E (IS17218) sterility sources were included into the crossing scheme. The comparative assessment of the selected valuable traits of the maternal varieties and the research into the pollens in the flowering period did not reveal any differences. However, the pollen cytologic evaluation proved the differences between the A3 Zheltozerno 10, A4 Zheltozerno 10, and 9E Zheltozerno 10 maternal lines in the number and the type of the pollen defective grains [13]. In addition, the typically colored pollen grains were present on the cytoplasm of A4 and 9E types in the pollen of the sterile analogs, while the plants remained sterile (Fig. 1). The scientific literature also

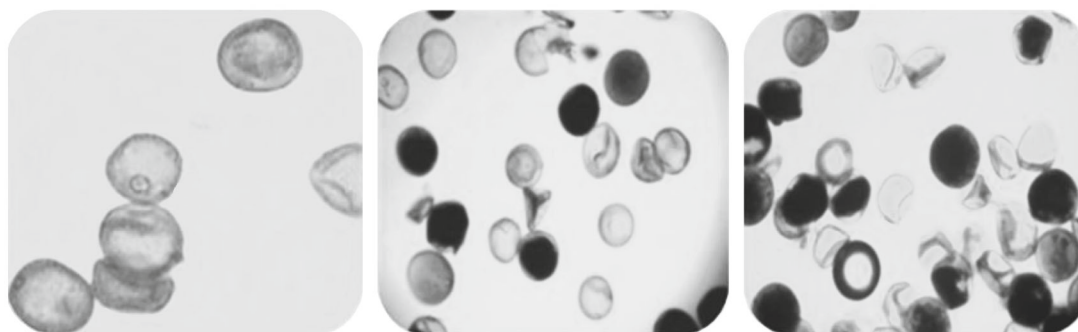


Fig. 1. Pollen of CMS lines A3 Zheltozernoe 10 (on the left), A4 Zheltozernoe 10 (in the middle), and 9E Zheltozernoe 10 (on the right).

describes the typical properties of the CMS-line pollen on the A4 and 9E cytoplasm [14].

The best grain sorghum lines bred in the Rossorgo Russian Research and Project-Technological Institute of Sorghum and Corn were used as the pollinators. At present, fifteen lines of them are acceptable for use in the arid regions of the Russian Federation, while three lines are undergoing government testing. These lines are as follows: Perspektivnyi 1 (L-1), Start (Sudzern 124), Merkurii (L-12), Ogonyok (Tonkostebelnoe 31), Kamelik (L-7-10), Topaz (L-4/07), Fakel (Norgum 06-10), Avans (20/2011), Azart (L-221-269/2010, Volzhskoe 615 (O-615), Geleofor (L-616), Kremovoe (AGS), Pishchevoe 614 (O-614), Sarmat (L-M Chernoplenchatoe 2), Vostorg (O-1237), Garant (ZhVI-875), Pishchevoe 35 (KP-35), and Magistr (L-KCI 28/13). The purity of the initial material in the experiment was annually maintained under the conditions of strict isolation.

The combining ability of the crossing components was determined with the topcross method [15]. The experimental data statistical processing was performed with the one-factor dispersion analysis using the *AGROS 2.09* software.

RESULTS AND DISCUSSION

One of the stages of the crossing component analysis is the assessment of the pollinator lines according to the responses to the CMS. Most pollinators included into this scheme are referred to the fixers of sterility of the A3, A4, and 9E types, excluding the Perspektivnyi 1 and Magistr lines ascertained to the A4 and 9E restorers of cytoplasm (Table 1).

A dispersion analysis of the selected valuable traits and the crop yields of the F_1 grain hybrids can indicate their significant differences ($F_{\text{fact.}} > F_{\text{theor.}}$), which pro-

Table 1. Response of paternal forms to A3, A4, and 9E CMS types, 2015–2016

No	Pollinator	CMS type		
		A3	A4	9E
1	Perspektivnyi 1	B	R	R
2	Ogonyok	B	B	B
3	Merkurii	B	B	B
4	Start	B	B	B
5	Kremovoe	B	B	B
6	Azart	B	B	B
7	Avans	B	B	B
8	Topaz	B	B	B
9	Volzhskoe 615	B	B	B
10	Pishchevoe 614	B	B	B
11	Pishchevoe 35	B	B	B
12	Sarmat	B	B	B
13	Kamelik	B	B	B
14	Geleofor	B	B	B
15	Fakel	B	B	B
16	Vostorg	B	B	B
17	Garant	B	B	B
18	Magistr	B	R	R

B—Sterility fixers; R—Fertility restorer.

Table 2. Assessment of F₁ hybrids for selection-valuable traits (cm) and crop yields (t/hectar), 2015–2017

Pollinator	CMS types of maternal lines																	
	Plant height			Inflorescence length			Inflorescence width			Labellum length			Labellum width			Grain yield		
	A3	A4	9E	A3	A4	9E	A3	A4	9E	A3	A4	9E	A3	A4	9E	A3	A4	9E
Perspektivnyi 1	178.4	168.3	169.9	19.9	19.1	18.9	9.4	10.7	9.6	61.7	56.3	54.3	5.0	4.9	4.7	3.34	4.52	3.87
Ogonyok	175.7	176.2	172.7	24.0	23.8	22.1	12.8	13.1	11.7	66.3	67.1	68.3	6.2	6.0	6.2	3.73	4.06	3.79
Merkurii	186.3	192.8	207.5	22.4	23.2	23.4	10.5	9.9	10.3	65.1	66.1	72.9	6.0	6.4	5.9	4.06	3.57	3.66
Start	180.5	183.5	197.8	21.3	21.5	21.4	8.9	8.9	9.4	60.0	63.8	64.1	4.9	5.3	5.2	5.59	5.51	5.41
Kremovoe	213.9	202.3	213.2	21.8	22.2	23.6	8.1	9.3	10.0	61.0	59.8	64.4	6.0	6.2	6.5	4.63	4.24	5.22
Azart	184.1	185.9	186.7	20.3	20.4	19.4	6.6	6.5	5.9	70.9	70.7	74.1	6.0	5.6	5.6	3.06	2.70	2.74
Avans	200.9	198.6	217.5	20.3	20.3	25.4	7.4	7.5	10.2	72.1	68.9	73.2	5.3	5.2	5.2	3.48	3.52	3.99
Topaz	185.7	183.2	177.6	21.6	21.6	22.3	7.7	7.7	7.7	77.1	74.1	71.9	6.6	6.8	6.8	3.45	3.01	3.24
Volzhskoe 615	183.4	178.9	165.7	20.1	18.9	22.5	10.5	9.2	12.6	66.7	62.2	57.6	5.2	5.4	5.3	5.13	4.26	4.58
Pishchevoe 614	192.9	201.8	202.6	20.3	21.4	21.9	7.5	7.9	8.5	71.9	67.2	73.2	5.4	5.8	6.0	4.37	4.34	4.07
Pishchevoe 35	199.6	192.7	197.7	20.1	19.7	19.8	6.8	7.3	7.3	67.5	67.9	68.3	5.1	6.1	6.2	4.08	3.29	3.47
Sarmat	194.5	186.5	193.9	22.5	20.9	22.8	9.6	7.7	8.9	70.4	67.6	66.8	5.3	5.2	5.2	4.75	4.72	4.80
Kamelik	159.6	154.8	170.7	20.2	20.1	22.0	7.5	8.5	8.6	66.8	66.1	68.6	5.3	5.4	5.7	3.09	2.72	3.31
Geleofor	197.7	188.5	191.2	17.9	17.6	19.3	7.2	6.9	7.2	73.9	71.9	71.9	5.2	6.0	5.8	2.59	2.44	2.51
Fakel	187.9	185.6	180.9	20.4	19.9	20.3	7.6	7.4	8.7	68.1	60.5	67.4	6.2	5.7	6.2	4.71	4.40	4.36
Vostorg	178.0	181.2	199.9	21.8	22.5	22.2	10.9	10.5	8.5	65.3	69.4	68.4	6.4	6.2	6.1	4.06	4.01	4.29
Garant	219.2	197.8	201.0	21.6	21.1	22.5	6.9	8.2	8.4	70.7	69.1	74.1	6.3	6.3	6.6	2.35	2.48	2.34
Magistr	176.8	177.2	175.9	18.0	19.9	21.6	6.7	7.2	10.7	62.8	66.5	73.0	5.5	6.2	6.5	3.38	3.17	3.71
F _{fact}	2.41*			1.99*			3.20*			2.86*			2.06*			2.14*		

**p* ≤ 0.05

Table 3. Combining ability of sorghum CMS lines by morphometric traits and grain crop, 2015–2017

CMS line	Plant height	Inflorescence		Labellum		Grain crop
		length	width	length	width	
GCA effects						
A3 Zheltozernoe 10	0.59	−0.31	−0.25	0.16	−0.12	0.06
A4 Zheltozernoe 10	−2.70	−0.32	−0.15	−1.12	0.03	−0.10
9E Zheltozernoe 10	2.11	0.63	0.40	0.95	0.09	0.03
F _{GCA} (CMS lines)	1.33	4.10*	2.44	2.35	1.52	0.39
SCA dispersion						
A3 Zheltozernoe 10	36.10	0.49	0.42	6.31	0.07	0.06
A4 Zheltozernoe 10	13.57	0.42	0.40	2.48	0.03	0.06
9E Zheltozernoe 10	46.40	1.13	0.92	6.87	0.04	0.04
F _{SCA} (CMS lines)	0.59	0.77	0.98	0.93	0.48	0.23

$p \leq 0.05$.

Table 4. SCA effects of the best crossing elements, 2015–2017

Crossing combination	Plant height	Inflorescence		Labellum		Grain crop
		length	width	length	width	
A3 Zheltozernoe 10/Perspectivnyi 1	5.61	0.91	−0.25	4.10	0.26	−0.63
A3 Zheltozernoe 10/Volzhscoe 615	6.81	−0.09	−0.02	4.37	0.02	0.41
A4 Zheltozernoe 10/Vostorg	2.17	−2.47	0.66	2.82	−0.07	12.54
9E Zheltozernoe 10/Merkurii	6.68	9.86	−0.23	3.913	−0.29	−2.60
9E Zheltozernoe 10/Kremovoe	4.88	1.29	0.43	1.71	0.18	18.97
9E Zheltozernoe 10/Avans	4.11	9.72	2.77	0.85	−0.12	−2.33
9E Zheltozernoe 10/Kamelik	−3.66	6.89	0.60	0.48	0.14	10.47
9E Zheltozernoe 10/Magistr	−4.49	2.84	1.13	4.61	0.34	40.63

vided the opportunity to study the CMS lines with the use of various types of the sterile cytoplasm by the combining ability (Table 2).

The analysis of the CMS-line combining ability with the Zheltozernoe 10 genome on the A3, A4, and 9E cytoplasm revealed a significant effect of a sterile cytoplasm on the inflorescence length. The 9E cytoplasm contributed to increasing the effects of the general combining ability (GCA), while the A3 and A4 cytoplasm contributed to decreasing its effects in the analyzed crossing scheme (Table 3). In addition, the higher GCA effects of the CMS-line Zheltozernoe 10 are recorded for the traits, such as the plant height (2.11), the inflorescence width (0.40), and the labellum parameters (0.09–0.96).

With respect to the specific combining ability (SCA) for the economically important traits, there was no significant difference between the A3 Zheltozernoe 10, A4 Zheltozernoe 10, and 9E Zheltozernoe 10 sterile lines. The highest SCA dispersion values for the plant height (46.40), the inflorescence parameters (0.92–1.13), and the labellum length (6.87) are recorded in the CMS lines with the 9E cytoplasm; the

lowest values for the grain yield were determined (0.04).

The genetic control of the analyzed traits proved that the additive genetic effects were dominant to the nonadditive effects in the A3, A4, and 9E CMS lines since the GCA and SCA mean square deviation ratio was represented as $ms_{GCA}/ms_{SCA} > 1$. The largest ratio of GCA to SCA mean square deviations was obtained for the *inflorescence length* trait.

The SCA effect analysis can provide the opportunity to allocate both the hybrid combinations with high heterosigosity and the initial material for inclusion into the three-line hybrid compound (Table 4). The fertile F₁ 9E Zheltozernoe 10/Magistr hybrid characterized by the low values for the SCA effects in the plant height (−4.49) and the high values for the parameters of inflorescence (1.13–2.84), labellum (0.34–4.61), and grain yield (40.63) was selected. This hybrid should be included into the competitive variety trial to assess its suitability for the mono feed and the grain forages.

The sterile F₁ hybrids may be used as the initial material in order to produce a three-line high-yielding hybrid suitable for grain and silage. These hybrids are as follows: 9E Zheltozerno 10/Kremovoe (for large and mean SCA effects on the plant height at growing, the inflorescence and labellum parameters, and the grain yield), 9E Zheltozerno 10/Merkurii (the plant height and the inflorescence and labellum lengths) with the large SCA effects on two or three traits, A3 Zheltozerno 10 in crosses with Volzhskoe 615 and Perspektivnyi 1 (the plant height and the labellum length), A4 Zheltozerno 10/Vostorg (the labellum length and the grain yield), 9E Zheltozerno 10/Avans (the plant height and the inflorescence parameters), 9E Zheltozerno 10/Kamelik (the inflorescence length and the grain yield).

CONCLUSIONS

Therefore, a significant effect of the sterile cytoplasm of the 9E type on the general combining ability of the sorghum CMS lines for the trait of the inflorescence length, which comprised 0.63, was revealed. Such a length was 0.32 (0.31) less for the compared A3 and A4 types. The traits (plant height, inflorescence and labellum parameters, and grain yields) occurring in the maternal lines of A3, A4, and 9E Zheltozerno 10 are controlled by the genes having an additive effect. The 9E Zheltozerno 10/Magistr hybrid is selected for the consequent competitive variety trials. With respect to producing the three-line high-yielding hybrids, seven sterile F₁ hybrids used as the initial material should be included. The results of assessing the combining capacity of the CMS lines (different only in the type of cytoplasm) can prove that the 9E type of sterility is preferable for use in breeding the sorghum with the improved economically valuable traits under the arid conditions of the southeastern part of Russia.

COMPLIANCE WITH ETHICAL STANDARDS

The authors declare that they have no conflict of interest. This article does not contain any studies involving animals or human participants performed by any of the authors.

REFERENCES

1. Crosatti, C., Quansah, L., Maré, C., et al., Cytoplasmic genome substitution in wheat affects the nuclear-cytoplasmic cross-talk leading to transcript and metabolite alterations, *BMC Genomics*, 2013, vol. 14, p. 868.
2. Reddy, B.V.S., Ramesh, S., Reddy, P.S., and Kumar, A.A., Male-sterility inducing cytoplasmic effect on combining ability in sorghum (*Sorghum bicolor* (L.) Moench),

Indian J. Genet. Plant Breed., 2009, vol. 69, no. 3, pp. 199–204.

3. Tyagi, V. and Dhillon, S.K., Cytoplasmic effects on combining ability for agronomic traits in sunflower under different irrigation regimes, *SABRAO J. Breed. Genet.*, 2016, vol. 48, no. 3, pp. 295–308.
4. Wei, W.L., Wang, H.Z., and Liu, G.H., Cytological and molecular characterization of a new cytoplasmic male sterility in rapeseed, *Plant Breed.*, 2009, vol. 128, no. 4, pp. 426–428.
5. Saxena, K.B., Sultana, R., and Mallikarjuna, N., Male-sterility systems in pigeon pea and their role in enhancing yield, *Plant Breed.*, 2010, vol. 129, no. 2, pp. 125–134.
6. Engelke, T., Hülsmann, S., and Tatlioglu, T., A comparative study of microsporogenesis and anther wall development in different types of genetic and cytoplasmic male sterilities in chives, *Plant Breed.*, 2002, vol. 121, no. 3, pp. 254–258.
7. Nothnagel, T., Straka, P., and Linke, B., Male sterility in populations of *Daucus* and the development of alloplasmic male-sterile line of carrot, *Plan Breed.*, 2000, vol. 119, no. 2, pp. 145–152.
8. Young, J.B. and Virmani, S.S., Effects of cytoplasm on heterosis and combining ability for agronomic traits in rice (*Oryza sativa* L.), *Euphytica*, 1990, vol. 48, pp. 177–188.
9. Ekiz, N., Safi Kiral, A., Akcin, A., and Simsek, L., Cytoplasmic effects on quality traits of bread wheat (*Triticum aestivum* L.), *Euphytica*, 1998, vol. 100, nos. 1–3, pp. 189–196.
10. Chandra-Shekara, A.C., Prasanna, B.M., Singh, B.B., Unnikrishnan, K.V., and Seetharam, A., Effect of cytoplasm and cytoplasm-nuclear interaction on combining ability and heterosis for agronomic traits in pearl millet (*Pennisetum glaucum* (L.) Br.R.), *Euphytica*, 2007, vol. 153, nos. 1–2, pp. 15–26.
11. Reddy, B.V.S., Ramesh, S., Reddy, P.S., and Kumar, A.A., Male-sterility inducing cytoplasmic effect on combining ability in sorghum (*Sorghum bicolor* (L.) Moench), *Indian J. Genet. Plant Breed.*, 2009, vol. 69, no. 3, pp. 199–204.
12. *Metodika gosudarstvennogo sortoispytaniya sel'skokhozyaistvennykh kul'tur* (Methods of State Variety Testing of Agricultural Crops), Moscow, 1985, vol. 2.
13. Kibal'nik, O.P., Selection value of new CMS types in sorghum, *Cand. Sci. (Biol.) Dissertation*, Saratov, 2009, pp. 43–62.
14. Tsvetova, M.I. and El'konin, L.A., Cytological study of the male generative sphere in CMS-lines of sorghum with type 9E cytoplasm, *Vestn. Samar. Gos. Agrarn. Univ.*, 2007, suppl., pp. 102–106.
15. Savchenko, V.K., A method for assessing the combinational capacity of genetically heterogeneous sets of parental forms, in *Metodiki genetiko-seleksionnogo i geneticheskogo eksperimentov* (Methods of Genetic Breeding and Genetic Experiments), Minsk, 1973, pp. 48–77.

Translated by O. Zhiryakova