Vol. 13(45), pp. 2610-2617, 8 November, 2018
DOI: 10.5897/AJAR2018.13381
Article Number: FE2FCCE59238
ISSN: 1991-637X
Copyright ©2018
Author(s) retain the copyright of this article
http://www.academicjournals.org/AJAR

ACADEMIC JOURNALS
expand your knowledge

African Journal of Agricultural Research

Full Length Research Paper

Influence of cytoplasmic genetic male sterility in the grain yield of maize hybrids

Luiz Augusto Rodrigues[1], Renzo Garcia Von Pinho[2*], Luiz Antonio Yanes Bernardo Júnior[3], Vander Fillipe de Souza[4], Francielly de Cássia Pereira[3], André Humberto de Brito[1] and Edmir Marques Faria Filho[1]

[1]Corteva Agriscience, AMG900-1105 Road, Uberlândia to Indianópolis, km 06, Zip Code 38490-000, Indianópolis, MG, Brazil.
[2]Agriculture Department, Federal University of Lavras-UFLA, Doutor Sylvio Menicucci Ave, 1001, Presidente Kennedy, P. O. Box 3037, Zip code 37200-000, Lavras, MG, Brazil.
[3]Biology Department, Federal University of Lavras-UFLA, Doutor Sylvio Menicucci Ave, 1001, Presidente Kennedy, P. O. Box 3037, Zip code 37200-000, Lavras, MG, Brazil.
[4]Agriculture Department, Federal University of Lavras-UFLA, Doutor Sylvio Menicucci Ave, 1001, Presidente Kennedy, P. O. Box 3037, Zip code 37200-000, Lavras, MG, Brasil.

Received 9 July, 2018; Accepted 10 September, 2018

One of the main barriers to the production of hybrid maize seeds consists of needing detasseling of female parent in order to avoid contamination with unwanted pollen. This activity is a laborious practice, which makes the production more expensive and promotes productive losses. Thus, the use of cytoplasmic genetic male sterility can facilitate the hybridization process, and consequently, the production of hybrid seeds. The objective of this study was compare grain yield in maize hybrids produced from the combination of two testers with five distinct lines of cytoplasm C "Charrua" versus the isogenic of fertile cytoplasm. Twenty common hybrids were evaluated in relation to grain crops and resistance to leaf diseases, white spot and gray leaf spot. The experiments were carried out during summer season of 2015/2016 and winter season of 2016. The use of lines with cytoplasm C was observed and it is promising since it did not affect the agronomic performance of hybrids, regardless of environment, crop season, parental and tester used. Therefore, cytoplasm C can be used as an excellent source of cytoplasmic genetic sterility in the production of hybrid seeds in seed companies that want to decrease the use of detasseling practice.

Key words: *Zea mays* L, production of seeds, detasseling.

INTRODUCTION

Maize is one of the most cultivated cereals in the world. In Brazil alone, more than 16 million hectares were sown during summer 2017/2018 and winter 2018 seasons. The average Brazilian corn yield is over 5 kg.ha^{-1} (Conab,

---

*Corresponding author. E-mail: renzovonpinho@gmail.com.

2018), and this is mainly because of hybrid seed adoption and management technologies. The simple hybrids ensure greater uniformity and enables high grain yield. However, the production process of hybrid seeds shows complications, such as the necessity of detasseling of female plants (Magalhães et al., 1999).

The manual corn detasseling is a seasonal and expensive practice. Consequently, this practice promotes an increase in production cost of hybrid seeds. Although there is equipment that enables the mechanization of this process, it is essentially manual repass. An alternative to eliminating labor in this process is the use of male sterile lines (Magalhães et al., 1999). The use of cytoplasmatic male sterility (CMS) and restoration fertility through nuclear genes permit the commercial exploitation of CMS systems for the production of hybrid seeds through the elimination of manual detasseling operation and generation assurance, F_1 fertility (Schnable and Wise, 1998). CMS is induced by the complementary action of nuclear and cytoplasmatic genes. These specific mutations in mitochondrial DNA make the plant produce no pollen or produce pollen that is not functional. Nevertheless, female fertility is not influenced by CMS, that is, male sterile plants can produce seeds if there is viable pollen available (Weider et al., 2009).

Basically, the CMS of maize can be divided into groups: CMS-T (Texas), which was practically eliminated in seed production due to the vulnerability to the fungus *Helminthosporium maydis* (Tatum, 1971; Duvick, 1973); CMS-S (USDA), which is unsteady and frequently shows fertility restoration; and CMS-C (Charrua), which has steady male sterility and a positive effect on grain yield (Weider et al., 2009; Stevanovic et al., 2016). Several types of research were realized in the area of molecular biology and cytogenetics with cytoplasm C (Lu et al., 2010; Yongming et al., 2016; Chen et al., 2016); however, information about the stability and productivity of materials in different environmental conditions is still very little disseminated (Weider et al., 2009), especially in tropical countries, such as Brazil. Considering this, the objective of this work is to evaluate agronomic performance of hybrids with cytoplasm C versus isogenic of normal cytoplasm from the main heterotic groups used by private breeding programs.

MATERIALS AND METHODS

The trials were carried out in the research stations of the company Dow AgroSciences in Brazil, in the summer season of 2015/2016 and in the winter season of 2016. During the summer season, the evaluations were carried out in four environments under irrigated conditions (Iguatama-MG, Indianópolis(1)-MG, Perdizes-MG and Varjão de Minas-MG) and seven non-irrigated conditions (Guarda Mor-MG, Indianópolis(2)-MG, Padre Olegário-MG, Uberaba-MG, Cascavel-PR, Castro-PR, and Itararé-SP). In the winter season, the trials were carried out in 13 environments under non-irrigated conditions (Montividiu-GO, Araguari-MG, Indianópolis-MG, Nova Mutum-MT, Primavera de Leste-MT, Sapezal-MT, Sorriso-MT, Cambé-PR, Cascavel-PR, Palotina-PR, Santa Terezinha do Itaipú-PR, Guaíra-SP, and Maracaí-SP).

The experimental design used was randomized blocks with two replicates. The plots were made up of four rows measuring four meters with spacing of 0.5 m between lines. At the moment of the sowing, NPK formulation used was 09-29-09 in doses of 450and 300 kg.ha^{-1} for summer and winter seasons, respectively. The application of nitrogenous and potassic fertilizers were incorporated through sowing. In general, the dose of urea varied from 200 to 350 kg.ha^{-1}, and the dose of potassium chloride varied from 100 to 150 kg.ha^{-1}, distributed twice during the crop cycle. The crop management, such as pest control, weeds and diseases were made according to the recommendations for maize.

Twenty treatments were evaluated, being 10 simple sterile hybrids of cytoplasm C and their respective iso-hybrid of fertile cytoplasm, from the crossings of five sterile lines of cytoplasm C with two testers. The female genitor lines belonged to the heterotic groups Tuxpeno, Suwan and Cateto, while testers belonged to groups Cateto and Suwan. These testers, in combination with lines of cytoplasm C, produced hybrids partially and totally fertile, respectively. The seeds of 20 simple hybrids were produced by manual crossing in pollination fields of research station in Indianópolis during the winter season of 2015. After the flowering, the foliar diseases, white spot (*Phaeosphaeria maydis*/ *Pantoea ananas*) and gray leaf spot (*Cercospora zeae-maydis*), were evaluated through a diagrammatic scale proposed by Agroceres (1996). In this scale, score 1 represents plots with 100% foliar tissue undermined or premature death, and score 9 represents plots with no foliar spots. As the incidence of the pathogens is dependent on specific conditions, the environments chosen for the evaluations were those that presented greater occurrences of foliar diseases. The weight of grains in each plot was evaluated in the moment of harvest and submitted to the variance analysis according to the following statistical models (1):

$$y_{ijk} = \mu + b_i + h_j + l_k + hl_{jk} + e_{ijk} \tag{1}$$

y_{ijk}: value observed of the plot that received hybrid i of block j in the environment k;
μ: general average associated with all observations;
b_i: effect of i-th block, being b_i ~N $(0, \sigma_b^2)$ (i = 1 and 2);
h_j: effect of j-th hybrid, being h_j ~N $(0,\sigma_h^2)$ (j = 1, 2, 3, ... 10);
l_k: effect of k-th environment, being l_k ~N $(0,\sigma_l^2)$ (k = 1, 2, 3, … 11 for summer season; k = 1, 2, 3, … 13 for winter season; and k = 1, 2, 3, ... 24 for joint analysis);
hl_{jk}: effect of interaction between hybrids and environments;
e_{ijk}: average experimental error associated with plot ij, being e_{ij} ~N $(0, \sigma_e^2)$.

In the variance analysis, the decomposition of hybrid effect on sterile lines, fertile lines and testers was realized. The averages were compared through mean contrasts, using Scott-Knott test (Scott and Knott, 1974).The experimental accuracy (2) per environment was calculated according to Resende and Duarte (2007):

$$\hat{r}_{gg} = \left[\frac{1}{1+(\sigma_e^2/b)/\sigma_h^2}\right]^{1/2} \tag{2}$$

σ_h^2: variance between hybrids; σ_e^2: residual variance; b: number of blocks in the environment.
All statistical analyses were realized in the software R (R Core Team, 2017).

RESULTS AND DISCUSSION

The summary of joint variance analysis for grain yield of the 20 hybrids evaluated is shown in Table 1. The sources of variation hybrids, environments and interaction hybrids x environments differ between them at significance level of 0.01. With detasseling of hybrids effect, it was also possible to observe occurring significant differences ($p < 0.01$) for sterile lines, fertile lines, testers and the interaction between lines and testers. Moreover, all interaction unfolding between hybrids and environments were significant. Significant differences ($p \geq 0.05$) were not observed for the contrast between sterile and fertile lines and for the contrast between testers and lines. Thus, it is possible to conclude fertile lines do not have grain yield that is higher than those of sterile lines, independent of the tester used.

The focus of this work is on verifying the possible effect of cytoplasm Charrua or CMS-C in the productive performance of hybrids and if they differ between environments and/ or with the tester used. Although the effect of hybrids was significant ($p < 0.01$), no significant differences was determined in the contrast of average grain yield of hybrids for cytoplasm C and version of fertile cytoplasm with 9.68 and 9.71 $t.ha^{-1}$, respectively.

While having a key role in respiration and energy production, mitochondria is also related to cytoplasmatic male sterility. However, most parts of its performance and role arise from genetic information found in the cell nucleus. That is because the number of genes in the mitochondrial DNA is very small. Though the mitochondrial maize genome is one of the most complexes, there are 58 to 60 genes involved in different metabolic processes (Clifton et al., 2004; Chen and Liu, 2014).

Thus, in order for difference between hybrids on the basis of cytoplasm to occur, at first, there are two possibilities. One of them is that although mitochondria has a few genes, they could be influenced by genotype of the line used as recurring genitor or by some environmental factor, such as in the case of cytoplasm T and occurrence of *Bipolaris maydis*. The other possibility would be during backcrossing, substituting the nucleus of the donor line of cytoplasm for the nucleus of the commercial line. The transference would not occur in all crosses and, in this case, the donor line can cause differences. From the results obtained in this work, it is possible to infer that the cytoplasm C did not affect the agronomic performance of the hybrid, and the backcrossings realized for transferring the nucleus were efficient. However, as female fertility is not affected by the presence of cytoplam CMS, and male sterile plants can produce seeds if there is viable pollen available (Weider et al., 2009), it is expected yield does not change in plants that have cytoplasm CMS.

Robledo et al. (2013) evaluated the sterile and fertile versions of two maize hybrids submitted to four different management systems of detasseling in two different plant densities. The author determined there were no differences between fertile and male-sterile versions in grain yield. Sangoi and Salvador (1998) and Magalhães et al. (1999) did not observe differences in grains production between both versions of genotypes evaluated. Table 2 summarizes the individual variance analysis for grain yield in 24 environments during summer of 2015/2016, and the second winter of 2016. With the exception of Araguari – MG, the contrast between fertile and sterile lines, and Primavera do Leste – MT, for the interaction contrast between lines and testers, the environments showed p-value as non-significant ($p \geq 0.05$).

Considering all environments for both seasons, grain yield varied from 3.15 to 13.65 $t.ha^{-1}$. During summer of 2015/16, the average yield was 11.86 $t.ha^{-1}$; while the average was 7.76 $t.ha^{-1}$ during winter. The trial in Uberaba - MG had the greatest yield in summer; while the trial in Santa Terezinha do Itaipu - PR had the greatest yield in winter, exceeding 10 $t.ha^{-1}$. On the other hand, Presidente Olegário - MG and Primavera do Leste - MT were the least productive trials during summer and winter, respectively. The experimental accuracy ($\hat{r}_{gg}$) vary from 0.81 to 0.97 (Table 2), which shows high experimental quality (Resende and Duarte, 2007).

During summer of 2015/2016, the hybrids were classified into four distinct groups. Hybrids H 01, H 04, H 06, H 07 and H 09 were the most productive with average varying from 12.63 to 13.89 $t.ha^{-1}$. Hybrids H 02, H 03 and H 10 were in the intermediate groups, with averages varying from 10.6 to 12.3 $t.ha^{-1}$. Finally, hybrid H 08 was the least productive, with average varying from 4.8 to 5.1 $t.ha^{-1}$. Only hybrid H 05 showed significant statistical difference between the versions; being the version of fertile cytoplasm higher than version CMS-C, based on the Scott-Knott test at 5% of probability (Table 3).

In the winter season, hybrids were classified in five distinct groups. The hybrids H 01 and H 04 were the most productive with the average varying from 8.8 to 9.2 $t.ha^{-1}$ and were followed by the hybrids H 03, H 06 and H 07 whose averages vary from 8.2 to 8.7 $t.ha^{-1}$. In the third and fourth groups were the hybrids H 10 and H 02, respectively. Finally, the hybrid H 08 was the least productive with average varying from 2.61 to 2.67 $t.ha^{-1}$. Only the hybrids H 05 and H 09 showed significant statistical differences between the versions of fertile cytoplasm and CMS-C, according to the Scott-Knott test at 5% of probability (Table 3).

It should also be noted that the climate conditions in the summer were favorable for the development of the crop even with no irrigation. During winter, low rainfall rates were registered. There were productivity losses in a large part of the producing regions due to the shortage of rain, mainly in the Southeast and Central West part of the

Table 1. Summary of joint variance analysis for grains yield (t.ha^{-1}) of 20 simple maize hybrids evaluated in 24 environments. The degrees of freedom (DF), mean square (MS) and p-value are shown for each source of variation.

Sources of variation			DF	MS	p-value
Blocks/ Environments (B)			24	1.32	0.52
Environments (E)			23	281.18	0.00
Hybrids (H)			19	220.24	0.00
	Lines (L)		9	232.46	0.00
		Sterile Line (SL)	4	276.50	0.00
		Fertile Line (FL)	4	247.00	0.00
		SL vs FL	1	0.30	0.63
	Tester (T)		1	271.16	0.00
	T x L		9	202.38	0.00
		T x SL	4	247.50	0.00
		T x FL	4	207.25	0.00
		T x (SL vs FL)	1	2.70	0.16
E x H			426	2.95	0.00
	E x L		206	3.23	0.00
	E x T		23	7.25	0.00
	E x L x T		197	2.14	0.00
error			441	1.37	

Table 2. Summary of the mean square (MS) and p-value of the decomposition of the effect of hybrids in sterile line (SL), fertile line (FL) and tester (T), grain yield (t.ha^{-1}) and experimental accuracy ($\hat{r}_{gg}$) of each of the 24 evaluated environments and repective havest season, summer of 2015/2016 and winter of 2016.

Environment	Season	SL vs FL		T (SL vs FL)		Grain yield (t.ha^{-1})	$\hat{r}_{gg}$
		MS	p-value	MS	p-value		
Padre Olegário-MG	Summer	7.489	0.064	0.167	0.772	9.408	0.95
Guarda Mor-MG	Summer	0.720	0.524	0.940	0.465	10.685	0.95
Perdizes-MG	Summer	0.802	0.602	4.276	0.235	10.987	0.88
Indianápolis(1)-MG	Summer	0.267	0.699	0.227	0.721	11.446	0.94
Cascavel-PR	Summer	2.678	0.119	0.083	0.777	11.695	0.97
Indianápolis(2)-MG	Summer	1.508	0.408	0.374	0.678	11.847	0.92
Itararé-SP	Summer	0.280	0.542	0.000	0.956	11.962	0.96
Castro-PR	Summer	1.957	0.162	1.540	0.212	12.739	0.97
Iguatama-MG	Summer	0.920	0.604	0.540	0.691	12.901	0.93
Varjão de Minas-MG	Summer	0.886	0.445	1.622	0.304	13.240	0.94
Uberaba-MG	Summer	0.087	0.787	1.375	0.290	13.645	0.97
Primavera do Leste-MT	Winter	0.039	0.700	2.619	0.006	3.153	0.88
Araguari-MG	Winter	5.024	0.021	0.125	0.697	5.268	0.93
Nova Mutum-MT	Winter	1.466	0.099	0.272	0.461	6.017	0.95
Maracaí-SP	Winter	0.011	0.895	1.148	0.192	6.336	0.92
Indianópolis-MG	Winter	0.045	0.847	1.683	0.245	7.518	0.93
Cambé-PR	Winter	0.084	0.815	0.444	0.591	7.720	0.89
Sorriso-MT	Winter	0.088	0.661	0.700	0.224	7.848	0.96
Palotina-PR	Winter	0.271	0.608	0.041	0.840	8.092	0.96
Montividiu-GO	Winter	2.243	0.205	0.049	0.848	8.559	0.90
Sapezal-MT	Winter	4.223	0.151	5.998	0.091	8.996	0.81
Cascavel-PR	Winter	1.754	0.278	0.371	0.613	10.301	0.92
Guaíra-SP	Winter	3.089	0.204	0.030	0.899	10.482	0.95
Sta T. de Itaipu-PR	Winter	4.740	0.062	3.550	0.103	10.679	0.98

Table 3. Grain yield (t.ha^{-1}) of 20 simple hybrids from five fertile and five male-sterile lines crosses with two testers in the summer season of 2015/16 and in the winter season of 2016.

Hybrid	Female	Male (Tester)	Grain yield (t.ha^{-1}) Summer 2015/2016		Winter 2016	
H 01	F66PW	M51	13.607	A	9.017	A
H 01C	F66CPW	M51	13.894	A	9.103	A
H 02	F16PW	M51	11.305	C	6.832	D
H 02C	F16CPW	M51	11.505	C	6.809	D
H 03	F20PW	M51	12.375	B	8.515	B
H 03C	F20CPW	M51	12.093	B	8.268	B
H 04	F25PW	M51	13.289	A	8.813	A
H 04C	F25CPW	M51	12.960	A	9.144	A
H 05	F21PW	M51	12.638	A	8.335	B
H 05C	F21CPW	M51	11.927	B	7.920	C
H 06	F66PW	M52	13.398	A	8.523	B
H 06C	F66CPW	M52	13.162	A	8.689	B
H 07	F16PW	M52	12.903	A	8.336	B
H 07C	F16CPW	M52	13.604	A	8.747	B
H 08	F20PW	M52	5.151	D	2.675	E
H 08C	F20CPW	M52	4.851	D	2.610	E
H 09	F25PW	M52	13.438	A	8.721	B
H 09C	F25CPW	M52	13.629	A	9.203	A
H 10	F21PW	M52	10.619	C	7.688	C
H 10C	F21CPW	M52	10.972	C	7.870	C
Hybrids of Fertile Lines (PW)			11.872	A	7.746	A
Hybrids of Male-sterile Lines (CPW)			11.860	A	7.836	A

PW female fertile; CPW female sterile. Averages vertically followed by the same letter do not differ statistically by the Scott-Knott test at 5% of probability.

country. As a result of this, the yield of the trials varied from 3.15 to 10.6 t.ha^{-1}**.**

Table 4 shows the grain yield based on the parents. The results indicate there were significant differences between the hybrids obtained by each tester. The hybrids obtained from M51 were significantly higher, with an average yield of 10.28 t.ha^{-1}; while the hybrids of M52 produced 9.16 t.ha^{-1}. For crossings done with the tester M51, hybrids obtained from lines F25CPW, F25PW, F66CPW, and F66PW were the most productive and did not have significant differences between them. The least productive hybrids were obtained from lines F16CPW and F16PW with yield lower than 9 t.ha^{-1}. For crossings done with the tester M52, the most productive hybrids were those obtained from lines F16CPW, F25CPW and F25PW. The hybrids that had the worst performance were those obtained from crossing with lines F20CPW and F20PW.

According to the Scott-Knott test at 5% of probability, out of ten hybrids tested in both versions, only three showed significant differences in grain yield. Even so, F20PW/M51, F21PW/M51, and F16PW/M52 had their yields very close to the version CMS-C. Although some hybrids showed significant differences between versions of fertile cytoplasm and CMS-C, in the set of hybrids this difference is approximately 40 kg.ha^{-1}.

Historically, the use of male-sterile cytoplasm type T was highly criticized because of susceptibility to *B. maydis* (Tatum, 1971). Thus, the aim of this work was to evaluate the main diseases whose occurrence and severity are common in this region during summer and winter. Nevertheless, the occurrence of the disease depends on the presence of pathogens, a host that provides low genetic resistance and climate conditions favorable for the development of the pathogen; consequently, the study was limited to specific locals and conditions. The evaluations of foliar diseases white spot and gray leaf spot, according to Agroceres scale (1996) adapted, is in Table 5. The evaluations were realized in Irai de Minas – MG, during summer of 2015/2016; and in Indianópolis – MG, during winter of 2016, whose environments were more favorable to the evaluation due to the occurrence of pathogens. The use of fungicides associated with the usage of hybrids adapted that provide good tolerance to diseases did not permit the emergence and development of other pathogens, such as *Puccinia*

Table 4. Mean grain yield (t.ha^{-1}) based on the combinations between sterile lines (CPW at the end of the name) and fertile lines (PW at the end of the name) with each tester (M51 and M52).

Lines	Testers				
	M51		M52		Average
F16CPW	8.960	D	10.920	A	9.930
F16PW	8.880	D	10.430	B	9.660
F20CPW	10.020	C	3.660	E	6.870
F20PW	10.440	B	3.940	E	7.230
F21CPW	9.760	C	9.260	D	9.510
F21PW	10.310	B	9.150	D	9.760
F25CPW	10.970	A	11.230	A	11.100
F25PW	11.270	A	10.880	A	11.060
F66CPW	11.300	A	10.740	B	11.020
F66PW	11.120	A	10.760	B	10.940
Sterile lines (CPW)	10.190	a	9.180	b	9.690
Fertile lines (PW)	10.370	a	9.140	b	9.730

Averages vertically followed by the same capital letter and horizontally by lowercase do not differ between them by the Scott-Knott test at 5% of probability.

Table 5. Scores[1] of diseases for white spot (*P. maydis*/ *P. ananas*) and gray leaf spot (*C. zeae-maydis*) evaluated in 20 single hybrids in two locals, Irai de Minas - MG in summer season of 2015/16 and Indianópolis - MG in the winter season of 2016.

Hybrid	Female	Male (Tester)	White spot		Gray leaf spot	
H 01	F66PW	M51	5	MS	7	R
H 01C	F66CPW	M51	4	MS	8	R
H 02	F16PW	M51	4	MS	6	MR
H 02C	F16CPW	M51	5	MS	7	R
H 03	F20PW	M51	6	MR	6	MR
H 03C	F20CPW	M51	6	MR	8	R
H 04	F25PW	M51	5	MS	7	R
H 04C	F25CPW	M51	5	MS	8	R
H 05	F21PW	M51	5	MS	7	R
H 05C	F21CPW	M51	5	MS	7	R
H 06	F66PW	M52	5	MS	7	R
H 06C	F66CPW	M52	5	MS	7	R
H 07	F16PW	M52	4	MS	4	MS
H 07C	F16CPW	M52	4	MS	4	MS
H 08	F20PW	M52	5	MS	4	MS
H 08C	F20CPW	M52	4	MS	4	MS
H 09	F25PW	M52	5	MS	8	R
H 09C	F25CPW	M52	6	MR	8	R
H 10	F21PW	M52	6	MR	5	MS
H 10C	F21CPW	M52	6	MR	3	S
Hybrids of fertile lines			5	MS	6	MR
Hybrids of Male-sterile lines			5	MS	6	MR

[1] Scale of foliar damage: 9 (0%) AR = highly resistant; 8 (1%) R = resistant; 7 (10%) R = resistant; 6 (20%) MR = moderately resistant; 5 (30%) MS = moderately susceptible; 4 (40%) MS = moderately susceptible; 3 (60%) S = susceptible; 2 (80%) S = susceptible; 1 (>80%) AS = highly susceptible.

spp., Exserohilum turcicum, and *B. maydis*. This enabled a good characterization for white spot and gray leaf spot. The scores of white spot varied from 4 to 6, that is, they vary from moderately susceptible to moderately resistant. The hybrids H 03, H 03C, H 09C, H 10 and H 10C were moderately resistant, and the others were classified as moderately susceptible. Only hybrid H 09 obtained distinct classification in relation to its CMS-C isogenic.

The white spot is commonly associated with the fungus *P. maydis* and there are several molecules of fungicides registered to control this disease in maize crop in Brazil (Agrofit, 2018). However, Paccola-Meirelles et al. (2001) isolated the bacterium *P. ananas* with high frequency. Since then, this bacterium has been considered the etiologic agent of the white spot because it was the only pathogen to be successfully submitted to Koch's postulates (Manerba et al., 2013). Therefore, the genetic resistance as tool to control the white spot is the most effective and low-cost strategy (Carson, 2001; Schuelter et al., 2003; Lana et al., 2017). In this study, there was a small variation in the response of hybrids in relation to the tolerance to white spot. This brings a low variability of genotypes tested back, since no hybrid had full resistant behavior. Generically, hybrids CMS-C did not worsen or improve the level of tolerance to white spot when compared to their isogenic of fertile cytoplasm. For gray leaf spot*,* scores varied from 3 to 8, and the hybrids were classified between susceptible and resistant. Although many genotypes showed good tolerance to this disease, no hybrid was classified as highly resistant. The hybrids H 01, H 04, H 05, H 06 and H 09 were classified as resistant and did not differ from versions CMS-C. On the other hand, hybrids H 07 and H 08 were classified as moderately susceptible and did not show difference from their CMS-C isogenic. The hybrids H02 and H03 were classified as moderately resistant, while their CMS-C versions were classified as resistant. Finally, the version of fertile cytoplasm of hybrid H 10 was classification as moderately susceptible, higher than its CMS-C version, classified as susceptible. Although the hybrids show higher scores for gray leaf spot, male sterile cytoplasm did not contribute to this condition.

In general, hybrids of fertile cytoplasm obtained the same performance as hybrids CMS-C. Although studies such as that of Calugar et al. (2018) demonstrate that some agronomic characteristics of maize hybrids may be influenced by the alteration of the cytoplasm of the maternal genotype and the interaction between the cytoplasm and the testers, no significant changes were observed in the grain yield between fertile strains and CMS-C in the present study. Despite these results, the interaction among other lines may cause different reactions in the yield of grains or other agronomic characteristics. Stevanovic et al. (2016) observed higher yields of C and S cytoplasm maize lines in relation to normal cytoplasm lines. The authors state that a positive effect of CMS cytoplasm is expected because, as there is no pollen formation, there is less energy and nutrient consumption. In addition, Stevanovic et al. (2016) also observed higher grain yield in C cytoplasm genotypes than in S cytoplasm. Therefore, C type is somewhat more suitable for seed production.

Conclusion

The use of cytoplasm CMS-C to avoid manual detasseling is promising since it does not affect the agronomic performance of the hybrid. The productivity of hybrids CMS-C showed no considerable differences in relation to the fertile isogenic hybrids, independently of the environment, season, parental whose cytoplasm was incorporated and tester used. In addition, the hybrids CMS-C do not differ in tolerance to white spot and gray leaf spot when compared to isogenic fertile cytoplasm. The results constitute additional stimulus for companies who want to adopt cytoplasm C as an efficient and safe strategy to reduce the practice of manual detasseling.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

ABBREVIATIONS

CMS, Cytoplasmic male sterility.

ACKNOWLEDGMENTS

The authors would like to thank Corteva Agriscience, Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES), Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) and Fundação de Amparo à Pesquisa do Estado de Minas Gerais (FAPEMIG) for their support.

REFERENCES

Agroceres (1996). Guia Agroceres de sanidade**.** São Paulo: Sementes Agroceres 72p.

Agrofit (2018). Sistema de Agrotóxicos Fitossanitários. Available at: < http://agrofit.agricultura.gov.br/agrofit_cons/principal_agrofit_cons >. Access: May, 16, 2018.

Calugar RE, Has VV, Varga A, Vana C D, Copandean A, Has I (2018). The role of cytoplasmatic diversification on some productivity traits of maize. Euphytica 214:90.

Carson ML (2001). Inheritance of resistance to Phaeosphaeria leaf spot of maize. Plant Disease 85(7):798-800.

Chen L, Liu Y (2014). Male sterility and fertility restoration in crops. Annual Review of Plant Biology 65(5):579-606.

Chen B, Zhang Y, Lu Y, Wang J, Zhang S, Lan H, Rong T, Cao M (2016). DNA methylation analysis of sterile and fertile CMS-C hybrids and their parents in maize. Journal of Plant Biochemistry and Biotechnology 25(1):3-11.

Clifton SW, Minx P, Fauron CMR, Gibson M, Allen JO, Sun H, Thompson M, Barbazuk WB, Kanuganti S, Tayloe C, Meyer L, Wilson RK, Newton KJ (2004). Sequence and comparative analysis of the maize NB mitochondrial genome. Plant Physiology 136(3):3486-3503.

Conab -Companhia Nacional de Abastecimento (2018). Grãos - Série Histórica. Available at: <https://portaldeinformacoes.conab.gov.br/index.php/safra-serie-historica-dashboard>. Access in May, 16, 2018.

Duvick DN (1973). Potential usefulness of new cytoplasmic male steriles and sterility systems. In: Proc Annu Corn Sor Res Conf.

Lana UGP, Souza IRP, Noda RW, Pastina MM, Magalhaes JV, Guimaraes CT (2017). Quantitative Trait Loci and Resistance Gene Analogs Associated with Maize White Spot Resistance. Plant Disease 101(1):200-208.

Lu Y, Liu Y, Wang J, Cao M, Rong T (2010). Variation and Patterns of DNA Methylation in Maize C-type CMS Lines and their Maintainers. Journal of Plant Biochemistry and Biotechnology 19(1):43-50.

Magalhães PC, Durães FOM, Oliveira AC, Gama EEG (1999). Efeitos de diferentes técnicas de despendoamento na produção de milho. *Scientia Agricola* 56(1):77-82

Manerba FC, Souza PE, Von Pinho RG, Dornelas GA, Monteiro FP (2013). Antibióticos no Controle da Mancha Branca do Milho. Comunicata Scientiae 4(4):361-367.

Paccola-Meirelles LD, Ferreira AS, Meirelles WF, Marriel IE, Casela CR (2001). Detection of a bacterium associated with a leaf spot disease of maize in Brazil. Journal of Phytopathology 149(5):275-279.

R Core Team (2017). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.

Resende MDV, Duarte JB (2007). Precisão e controle de qualidade em experimentos de avaliação de cultivares. Pesquisa Agropecuária Tropical 37(3):182-194.

Robledo MT, Calderón AE, Pastor VT, Escamilla IA, Islas EC, Fernández AT, Macías MS, Bernal RV, Montiel NOG, Caballero AP, González BZ (2013). Eliminación de espiga y hojas en progenitores androestériles y fértiles de los híbridos trilineales de maíz 'H-47' y 'H-49'. Revista Fitotecnia Mexicana 36(3):245-250.

Sangoi L, Salvador R J (1998). Effect of plant maize detasseling on grain yield, tolerance to high plant density and drought stress. Pesquisa Agropecuária Brasileira 33(5):677-684.

Schnable PS, Wise RP (1998). The molecular basis of cytoplasmic male sterility and fertility restoration. Trends in plant science 3(5):175-180.

Schuelter AR, Souza IRP, Tavares FF, Santos MX, Oliveira E, Guimarães CT (2003). Controle genético da resistência do milho à mancha por Phaeosphaeria. *Revista Brasileira de Milho e Sorgo* 2(1):80-86. http://dx.doi.org/10.18512/1980-6477/rbms.v2n01p%25p

Scott AJ, Knott M (1974). A cluster analysis method for grouping means in the analysis of variance. Biometrics 30(3):507-512.

Stevanovic M, Camdzija Z, Pavlov J, Markovic K, Vancetovic J, Mladenovic Drinic S, Filipovic M (2016). The Application of Protein Markers in Conversion of Maize Inbred Lines to The Cytoplasmic Male Sterility Basis. Genetika 48(2):691-698.

Tatum L A (1971). The southern corn leaf blight epidemic. *Science* 171(3976): 1113–1116. 10.1126/science.171.3976.1113

Weider C, Stamp P, Christov N, Hüsken A, Foueillassar X, Camp K, Munsch M (2009). Stability of Cytoplasmic Male Sterility in Maize under Different Environmental Conditions. *Crop Science* 49(1):77–84. 10.2135/cropsci2007.12.0694

Yongming L, Zhuofan Z, Yanli L, Chuan L, Jing W, Boxiao D, Bing L, Tao Q, Wenbing Z, Moju C (2016). A preliminary identification of *Rf*-A619*, a novel restorer gene for CMS-C in maize (*Zea mays* L.). Peer J 4:e2719.