Influence of Cytoplasmic Male Sterility on Grain Yield of the "Turda" Maize Hybrids under Different Environmental Conditions

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ABSTRACT

In recent decades, little attention has been paid to using cytoplasmic male sterility (CMS) in maize hybrid seed production. Cytoplasmic male sterility is important for seed production of maize hybrids because CMS seeds are relatively inexpensive. The trait (CMS) is reversible and can be restored to fertility in the presence of nuclear restorer genes (Rf genes). There is very little knowledge about the effect of these different CMS types on the grain yield of modern hybrids. The present study was carried out in 2021, 2022 and 2023 in the central part of Romania, to investigate the specific impact of CMS on grain yield and other agronomic traits. 10 singlecross hybrids were tested, four of which were recently registered. Generally, CMS cytoplasm significantly negatively affected the yield of the hybrids tested over the three years. In 7 of the 10 hybrids tested, CMS cytoplasm influenced the insignificant decrease in average yield. In the higher-yielding 2021, only two hybrids tested with cms-C cytoplasm had a higher yield than hybrids with isogenic fertile cytoplasm. In 2022 and 2023, when yields were significantly reduced, due to water and high temperature stress conditions, the yield of the four tested hybrids with cms-C cytoplasm was higher than isogenic fertile cytoplasm. Recently developed hybrids: Turda 2020 and SURO 11 had the highest average grain yields, and the difference in yield between hybrids with CMS cytoplasm and those with isogenic fertile cytoplasm was non-significant. Therefore, for some hybrids in which the differences in grain yield between the CMS cytoplasm and the normal type were insignificant, in this case, we recommend the production of maize hybrid seeds based on the CMS cytoplasm.

Keywords: cytoplasmic male sterility, maize, grain yield, detasseling.

INTRODUCTION

For a long time, little attention has been paid to the increase in the maize yield (Zea mays L.) due to cytoplasmic male sterility (CMS). Lately, interest is growing because CMS seed is relatively inexpensive.

The production of maize hybrid seeds requires growing the parental inbred lines in the alternative blocks, in an isolated plot maternal parental form, which must be detasseled before they have shed any pollen, including all the plants on which the seed will be produced.

This can be achieved by detasseling (manually or mechanically), applying chemicals that prevent pollen formation or its dispersal, and by using CMS. Manual detasseling is difficult to work and expensive. It requires large number of people who may work as little as one week to more than five weeks. Manual labour is the main issue in detasseling (Has, 2006; Jovanović et al., 2018; Rodrigues et al., 2018). Detasseling machines that cut or pull the tassels also destroy some leaves, reducing yield (Gheţe et al., 2020).

Cytoplasmic male sterility may be used to eliminate the detasseling, needed to produce hybrid seed. Nuclear restorer-of-fertility genes (Rf-restorer genes) restore male fertility in the first generation after crossing a CMS and a non-CMS plant carrying the restorer alleles (Rf). Three major types of cytoplasm that induce male sterility, have been defined in maize as cms-T, cms-S, and cms-C. Sources based on cms-T were abandoned in the early 1970s after an epidemic caused by the pathogen Hm-T, which was especially virulent in maize with cms-T cytoplasm (Has, 2002).

Recently, breeders in Europe and the USA have become again interested in CMS as a convenient and cost-effective tool. They

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recommended using mainly cytoplasmic male sterility of type cms-C (Stamp et al., 2000; Has, 2006; Jovanović et al., 2018; Rodrigues et al., 2018).

The system of restoration of cytoplasmic male-sterility cms-C is complex. Restorer genes are many throughout most gene pools, limiting the selection of maternal lines. Furthermore, partial restoration often occurs in maternal lines, which is expressed variably, in different environments (Duvick and Noble, 1978; Has et al., 1987; Saigon and Salvador, 1996; Weider et al., 2009). Partial restorations can lead to selfpollination of the maternal parent and, thus, to impure hybrid seeds. The cms-C stability is high in hot air temperatures, decreased in lower temperatures, and depends on the hybrid genotype (Buckmann et al., 2016).

As a result of maize studies, Ukrainian Prysiazhniuk breeders et al. (2020),determined that DNA markers to C and S types of sterility are a quick and reliable approach for identifying sterile maize lines, in contrast to field studies, which have several limitations (temperature, humidity, sowing dates, daylight hours). The use of DNA (PCR) with specific primers for C and S types of cytoplasm markers is useful for the determination of the difference between lines which are morphologically identical and differ only by the type of cytoplasm, and also to determine the types of CMS based hybrid.

Many investigations of the interaction between genotypes and male-sterile cytoplasm were carried out after 1975, mainly concerning grain yield (Cabulea et al., 1994; Has et al., 2002; Dhillon et al., 2008; Kohls, The cms-T cytoplasm 2010). continued to be used only in some hybrids cultivated in the northern areas of Romania, Hungary and Russia, where the ecological conditions are not favorable for the development of the pathogen (Nemet, 1981; Cabulea et al., 1987; Sarca 2004; Has, 2006).

A physical mixture of different CMS types was already under discussion in the early 1970s to reduce the risk of a new cytoplasmspecific pathogen such as Hm-T (Gracen and Grogan, 1974). According to Duvick and Noble (1978), there may be a concern that all

maize may again be of the same cms-S or cms-C type, giving rise to ways to prevent this narrowing of the base of the cytoplasmic gene. So, both Duvick and Noble (1978), as well as other breeders (Cabulea et al., 1994; Zeng et al., 1998; Sarca, 2004), propose the application of the term "multiplasm" in the technique of producing maize hybrid seeds that contain a mixture of several types of male sterile cytoplasm. They proposed that maternal lines be represented by two or more cytoplasmic male sterility sources, and the paternal form to be a common restorer. The genetic diversity of the cytoplasm prevents the unidirectional evolution of specialized races of pathogens (Cabulea et al., 1987). Some seed producers prefer to mix seed produced on CMS cytoplasm with that produced on male-fertile cytoplasm, even when fertility restorers are used (Cabulea et al., 1994; Sarca 2004). Robledo et al. (2018) approached this method and considered that the best mix of seed in practical terms would be the proportion of 80% male sterile and 20% fertile seed because it is more convenient to have the highest proportion of plants with male sterile that will not need detasseling during seed production.

Large-scale cultivation of maize hybrids based on a single source of CMS may pose a serious challenge to sustainable grain yield because of decreasing genetic diversity. There is an ongoing need to produce and evaluate the new hybrids on various CMS systems, evaluate various CMS systems in different genetic backgrounds for their effects on agronomic characteristics of the plant, and susceptibility to pests and diseases to develop strategies for large-scale deployment of the pest-resistant hybrids on farmer's fields. (Kaeser et al., 2003; Dhillon et al., 2008). The idea would be good, but the main difficulty in implementing it, is that we are not sure that we could have the possibility to maintain so many different forms of CMS that are available (Duvick and Noble, 1978; Calugar et al., 2016).

In the USA, the Seed Growers Association estimated that maize seeds sold in 1987 contained 66.1% normal cytoplasm, 22.1% cms-C and 11.5% cms-S seeds (Wych, 1988). Between 1994 and 2010, most maize hybrids developed at the Institute of Phytotechnology, Porumbeni, Republic of Moldova, and the maize seed production of these hybrids was based on CMS of type M. The area cultivated with maize hybridization lots based on CMS of type cms-M represents 71.5% of the area reserved for seed maize (Partas, 2011).

After the 1980s, the first hybrids produced on C-type male-sterile cytoplasm, created at research units in Romania, were developed at Fundulea and Turda. The results of experimentation of the hybrids produced on cms-C and cms-ES sources, compared to produced their analogues on normal cytoplasm, in several localities, did not show significant differences regarding the yield capacity (Sarca and Barbu, 1982; Sarca et al., 1985; Has et al., 1987; Sarca et al., 1990). The same results were obtained by Saigon and Salvador (1996), no difference in grain yield was found between sterile and fertile counterparts of the inbred and hybrid, regardless of plant population. In another experiment, the grain yield of the hybrids produced on the cms-C source was significantly lower than of their counterparts on normal cytoplasm (Has et al., 1987; Sarca 2004; Has, 2006; Oroian, 2010).

After experimenting with maize hybrids produced on three types of cytoplasm (cms-T, cms-ES, and fertile), tested under irrigated and non-irrigated conditions, Ionescu (2005) concludes that the conditions showed no significant differences between the two male sterility types and the fertile analogue, proving the opportunity of using the cms-ES cytoplasm type for producing the hybrid maize seed in our conditions.

The comparisons made at the Agricultural Research and Development Station (ARDS) Turda, Romania, between hybrids produced on cms-T and cms-C and their analogues with normal cytoplasm, showed that cms-C cytoplasm was significantly superior for grain yield compared to cms-T (Cabulea et al., 1994). The stability of the hybrids produced on cms-C was weaker than those on cms-T, regarding the grain yield depending on the specifics of the crossing formula (Has et al., 1987; Has, 2002; Oroian, 2010).

Based on the data obtained and the existing knowledge in the speciality literature, it is necessary to use several sources of cytoplasmic male-sterility, but only after studying, checking in different ecological conditions, and carefully choosing the most valuable ones for seed production (Duvick, 1965; Sarca et al., 1990; Has et al., 2002).

The present study was carried out in 2021, 2022, and 2023 in the central part of Romania, at ARDS Turda, to investigate the specific impact of CMS on grain yield and other agronomic traits, the effects of different maize cytoplasm types (normal and cms-C) as well as the interactions (cms x Rf) of male-sterile cytoplasm (cms-C) with hybrid genotype, of the recently registered maize hybrids and new crosses.

MATERIAL AND METHODS

Field trial research was conducted at the Agricultural Research and Development Station (ARDS) Turda, in the Transylvanian Plain, Romania. Some limiting factors are characteristic of the climatic conditions of the Transylvanian area during the maize growing season. Climatic conditions correspond to the FAO groups 250-300-380+, spring low-temperature, and shorter growing seasons (Has et al., 2022). In this area, the early and semi-early hybrids find good conditions for cultivation.

Experiment design. The experimental design consisted of randomised complete split-plot blocks (RCBs), with three factors: 1) year as a large plot with three graduations, 2) maize hybrids, with ten graduations as a medium plot, 3) cytoplasm type as small plots with two gradations: normal type (hybrid seed produced by detasseling) and cms-C type (hybrid seed produced by cms-C). The harvested area of each plot was of 9.8 m². Each plot was replicated three times.

No.	Hybrid	Registered year	No.	Hybrid	Registered year
1	Turda 248	2012	6	HST A 483 - 33	new
2	Turda 332	2014	7	HST A 483 - 39	new
3	Turda 2020	2021	8	HST A 483 - 38	new
4	SURO 11	new	9	HST A 483 - 15	new
5	HST A 483 - 2	new	10	HST C 344 - 427	new

Table 1. The ten single cross hybrids studied, developed and registered at ARDS Turda - Romania

To evaluate the performance of single cross hybrids, the Relative Selection Index (RSI) was calculated; based on the formula:

$$RSI \% = \frac{GYx(100 - GM)x(100 - BS)}{\overline{X}GY x \overline{X}(100 - GM) x \overline{X}(100 - BS)} x 100$$

RSI = Relative Selection Index (%); GY = Grain Yield (kg/ha); GM = Grain Moisture (%); BS = Broken stalks (%).

The value of the relative selection index was interpreted statistically considering 100% of the absolute value of the selection index corresponding to the average of the experience (Has et al, 1987).

Cultivation technology. The analysis of the experimental soil indicated that the dominant soils are chernozem, characteristic to the Transylvanian Plateau. As a chemical description, the soil has a weakly alkaline neutral pH, neutral to high humus content, well supplied in nitrogen and potassium, and medium supplied in phosphorus. The previous crop was wheat (Triticum aestivum, ssp. vulgare). 400 kg/ha of complex mineral fertilizer NPK 27:13.5:0 was applied at sowing. Weeds were controlled using glyphosate two weeks before land preparation, and Laudis 66 OD 2.0 l/ha postemergence.

The field trials were sown in the last decade of April (April 25 and 30), during 2021-2023. The trial was laid out in a split-plot design, with three replications. The planting density was 70,000 plants/ha. There were four rows in each sub-plot; the rows were 7.0 m in length and spaced 0.70 m apart. The hybrids were sown using the Wintersteiger seeder for experimental plots.

Biometrics were performed on 20 plants/plots, as well as on ears, at harvest.

Data recorded. The plants from the two central rows of each split plot were harvested, at the moment of technological maturity. Ears harvested from each plot were shelled and the grain moisture was determined using a (Model Granomat moisture tester PFEUFFER - GMBH - Made in Germany). Grain yield was adjusted to 14.0% grain moisture level. The following grain yield traits were measured using 20 random ears/plot, at harvest: grain yield (kg/ha); grain moisture content (%) and frequency of breaking plants at harvest (%).

Analysis of the variance (ANOVA) of the split-split plot design in RCB arrangement was performed based on individual plot observation using the method of Ceapoiu (1968). The significance of the factors was achieved by the difference from the mean of the factor, based on the degree of significance of the differences (LSD) after calculating the analysis of factors (variances) for a multifactorial experiment with split plots.

<u>Climatic conditions</u> (Figure 1). The meteorological data used were obtained from Turda Meteorological Station, longitude 23°47', latitude 46°35', altitude 427 m. The experimental results were influenced by some climatic peculiarities of the three years of field trial - 2021, 2022 and 2023.

The year 2021 was favorable for maize growing from the point of view of temperatures evolution; April and August may be appreciated as warm (+131.5°C compared to a normal multiannual of 62 years), and in terms of the rainfall regime, a slightly rainy year (+3.8 mm), dry conditions in June, and excessively rainy in July. The years 2022 and 2023 can be characterized as warm. In 2022, the rainfall regime (+30.8 mm) was normal for all maizegrowing season. However, June and July, considered two important months for achieving the harvest, were dry, while late August and September were rich in rain. These conditions led to an extension of the maize growing season and slowed the rate of grain water loss.

The year 2023 had poor water supply in May and the first decade of June, which influenced the uneven emergence of plants and a low rate of vegetative development. In 2023, the two months of August and September were rich in rain. These conditions influenced the extension of the growing season of maize and the reduction of the rate of water loss from the grains.

For maize grown in the Transylvanian Plain, the droughts in June (-42.8 mm) and July (-52.8 mm)/2022 (Figure 1) are more damaging because they coincide with a critical period for water stress, when reproductive organs appear, pollination takes place, the formation and filling of the grains is achieved, a fact also supported by Sarca (2004). In 2023, the drought occurred in May (-36.0 mm), when water would have been needed in the soil for the uniform germination of plants.

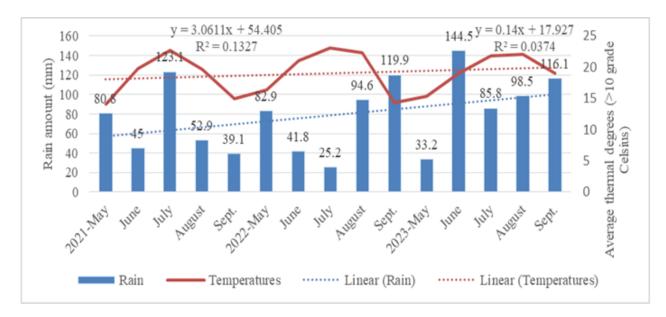


Figure 1. Climatic conditions during the period of maize growing season: average growing degrees and rainfall regime during the maize growing season, in Turda (2021-2023)

RESULTS AND DISCUSSION

Analyses of variance and genotype by environment interaction of grain yield and other agronomic traits. Table 2 presents the analysis of grain yield variants and some agronomic characters, in a polyfactorial experimental system with 10 hybrids on Ctype male-sterile cytoplasm and their fertile analogues, in 2021-2023. The experimental conditions (years) proved to be significantly different, ensuring diversified ecological verification conditions. For both grain yield, grain moisture at harvest and plant resistance to breaking distinctly significant differences were determined by the interaction of the two cytoplasms with the hybrid genotype.

Results of the combined analysis of variance (ANOVA) across the test environments of three years (Y), ten hybrids (H) and two types of cytoplasm (C) showed distinctly significant (p<0.01) mean squares for grain yield, grain moisture at harvest and frequency of broken stalks at harvest (Table 2). Additionally, significant (p<0.05) mean squares for Y × H, and H × C interactions were observed for grain yield and grain moisture at harvest. Similarly, significant (p<0.05) mean squares

for Y x H x C interaction were observed for grain moisture at harvest, except for the grain

yield, and frequency of broken stalks at harvest were not significant.

Source of	df	Grain yield kg/ha		Grain mois at harves (%)		Frequency of broken stalk at harvest (%)	
variance		s ² (Mean squares) Signif.	Rate of factor (%)	s ² (Mean squares) Signif.	Rate of factor (%)	s ² (Mean squares) Signif.	Rate of factor (%)
Year (Y)	2	371308000**	80.0	734.2**	72.8	674**	12.5
Error (Y)	4	213030	0.1	0.3	0.0		1.1
Hybrid (H)	9	8847664**	8.6	26.3**	11.7	489**	40.9
ҮхН	18	3194183**	6.2	12.5**	11.1	74**	12.5
Error (H)	54	242267	1.4	0.4	0.9		13.6
Cytoplasm (C)	1	6800113**	0.7	2.4*	0.1	162**	1.5
Y x C	2	340106	0.1	0.2	0.0	30	0.6
H x C	9	508909*	0.5	2.4**	1.1	31	2.6
Y x H x C	18	357284	0.1	1.1**	1.0	14	2.4
Error (C)	60	238507	1.5	0.4	1.2		12.0

Table 2. Combined analysis of variance across 2021 and 2023 years (% sum of squares)
of split plot design for studied 10 maize hybrids under two types of cytoplasm

<u>The effect of climate change</u>. There are many reasons to consider drought tolerance an important component of maize hybrids' success in drought-prone regions and years. In the zones relying on growing season rainfall (Turda-Romania), there is considerable inter-annual variation in the quantity of total rainfall and its distribution (Has et al., 2022; Simon et al., 2023), such as 2021-2023 (Figure 1).

In the favorable environmental conditions of the year 2021, three of the ten tested hybrids stood out for the highest grain yields (average of the two types of cytoplasm): Turda 2020 (14,751 kg/ha), Turda 332 (12,542 kg/ha), and SURO 11 (12,413).

In some years, grain yield can be significantly reduced by transient water limitations of varying critical timing, (Table 2). The ten hybrids tested on two different cytoplasm types, in the years 2022 and 2023, achieved lower grain yield, especially in 2022 the average yield was significantly lower (-2.410 kg/ha) compared to the year 2021, due to the water stress (Table 3). The climatic conditions in the Transylvanian Plain in 2022 (Simon et al., 2023), the droughts in June (-42.8 mm) and July (-52.8 mm) (Figure 1)

were damaging for maize because they coincided with a critical period for water: during the pollination period, the formation and filling of the grains (Has et al., 2022).

The experimental mean yield was higher in the first year, $12,074^{***}$ kg/ha compared to the second and third year (2022 and 2023) when the grain yields were $7,105^{000}$ kg/ha and $9,366^{ns}$ kg/ha, respectively (Table 3). Climatic changes during the three years, in which the ten hybrids produced on the two types of cytoplasm were tested, left their mark on the grain yield, because of the poor distribution of precipitation.

Nevertheless, considering the LSD of 798 kg/ha (for Y x H x C interactions) (Table 3), only in the case of hybrids SURO 11, HST A483-5, and HST A483-39, the differences in grain yield between the two types of cytoplasm (normal and cms-C) were significantly higher in 2022 (a year with dry summer). In the case of the recently registered hybrids Turda 248, Turda 332, and Turda 2020, the yield differences between the hybrids produced on normal cytoplasm and their analogues on male-sterile cytoplasm were non-significant, in all three years of the field trials. The average grain yield of the Turda 248 hybrid produced on male-sterile cytoplasm was higher compared to the fertile isogenic hybrid (Table 3). The significantly greater yield differences of HST A483-5 and HST A483-39 hybrids produced on normal cytoplasm compared to male-sterile cytoplasm analogues would not recommend these hybrids to be produced on male-sterile cytoplasm (cms-C).

Years	2021		2022		2023		Average	
Hybrid	Cytoplasm type	kg/ha	Signif. (Nrf-cms)	kg/ha	Signif. (Nrf-cms)	kg/ha	Signif. (Nrf-cms)	Signif. (Nrf-cms)
1.Turda 248	normal	10,456	-471 ^{ns}	6,342	+121 ^{ns}	8,743	-421 ^{ns}	-257 ^{ns}
	cms-C	10,927		6,221		9,164		
2.Turda 332	normal	12,633	+213 ^{ns}	7,253	+668 ^{ns}	9,209	-169 ^{ns}	+238 ^{ns}
	cms-C	12,420		6,585		9,378		
3.Turda 2020	normal	15,077	+653 ^{ns}	7,229	+361 ^{ns}	9,797	-177 ^{ns}	$+279^{ns}$
	cms-C	14,424		6,868		9,974		
4.SURO 11	normal	12,557	+289 ^{ns}	8,343	+1,225**	9,446	$+652^{ns}$	+722**
	cms-C	12,268		7,118		8,794		
5.HST A483 - 2	normal	12,125	+491 ^{ns}	7,336	-310 ^{ns}	10,160	+615 ^{ns}	+265 ^{ns}
	cms-C	11,634		7,646		9,545		
6.HST A483 - 33	normal	11,031	+77 ^{ns}	6,910	+956*	7,764	+324 ^{ns}	$+452^{ns}$
	cms-C	10,954		5,954		7,440		
7.HST A483 - 39	normal	13,221	+1,819***	7,989	+328 ^{ns}	10,695	+398 ^{ns}	+578*
	cms-C	11,402		7,661		10,297		
8.HST A483 - 38	normal	11,791	+852*	7,894	+280 ^{ns}	10,425	-202 ^{ns}	+310 ^{ns}
	cms-C	10,939		7,614		10,627		
9.HST A483-5	normal	12,184	+664 ^{ns}	7,545	+1,095**	10,288	+701 ^{ns}	+820***
	cms-C	11,520		6,450		9,587		
10.HST C344-427	normal	11,816	-282 ^{ns}	6,783	+417 ^{ns}	8,240	$+496^{ns}$	+210 ^{ns}
	cms-C	12,098		6,366		7,744		
Average - check	normal	12,289	+430**	7,362	+514***	9,477	+222 ^{ns}	+388***
	cms-C	11,859		6,848		9,255		
Average/y	12,074	+2,559***	7,105	$-2,410^{000}$	9,366	-149 ^{ns}	0	
LSD 0.05 Y x H x C = 798		Y x C =	= 252	$H \ge C = 460$) $C_1(Nr)$	$f) - C_2$ (cms	s-C) = 146	Y = 234
LSD 0,01 1061			335	612	2		194	388
LSD 0,001		436	797	7		252	726	

*, **, *** significant at 0.05, 0.01 and 0.001 probability levels, respectively; ns = non-significant; Nrf = normal cytoplasm.

Considering the significance of the interactions between the type of cytoplasm and the genotype of the hybrid for the phenotypic expression of the three characters, Table 4 draws attention to the fact that the general favorable or unfavorable effects of the cytoplasm can be intensified as an expression through the specific action with the genotype the hybrid, a fact supported by Cabulea et al. (1987), Has et al. (1989) and Cabulea et al. (1994).

The type of cytoplasm used to produce the studied hybrids, generally influenced both the vegetation period of the hybrids, by delaying

the hybrids with male-sterile cytoplasm of type C (Turda 332 and Turda 2020), as well as by improving the resistance to breakage at harvest in most of the hybrids (Table 4). The two recently registered hybrids Turda 332 and Turda 2020, produced on male-sterile cytoplasm of type cms-C, showed slight tendencies to decrease the grain moisture at harvest and improve the resistance of the lodging and broken stalks, while the yield capacity decreased non-significantly. Therefore, the use of cms-C cytoplasm to avoid manual detasselling is promising, because it does not affect the agronomic performance of the hybrids, similar results were recorded by Rodrigues et al. (2018).

To evaluate the performance of hybrids, a relative selection index (RSI) was used, to select the crossing formula, which simultaneously quantifies the grain yield, precocity, and resistance to lodging and breaking of the stalks (table 4). Hybrids: Turda 2020 (110-104%), Turda 332 (106-102%), HST A483-39 (110-102%) and HST A483-38 (106-103%) stood out for the highest selection index value (RSI), but also by the closest values between the two cytoplasm types on which the hybrids were produced.

Characte	Grain yield (GY)		Grain moisture at harvest (GM)		Frequency of broken stalk at harvest (BS)		RSI	
Hybrid	Cytoplasm type	kg/ha	Signif. (Nrf-cms)	%	Signif. (Nrf-cms)	%	Signif. (Nrf-cms)	%/average
1.Turda 248	normal	8,514	-257 ^{ns}	16,91	+0,63*	7,6	$-7,2^{00}$	87
	cms-C	8,771		16,28		14,8		93
2.Turda 332	normal	9,699	+238 ^{ns}	16,33	-0,19 ^{ns}	1,9	-0,9 ^{ns}	106
	cms-C	9,461		16,52		2,8		102
3.Turda 2020	normal	10,701	$+279^{ns}$	16,91	$-0,02^{ns}$	6,9	-3,0 ^{ns}	110
	cms-C	10,422		16,93		9,9		104
4.SURO 11	normal	10,115	+722**	18,81	+1,03**	5,3	$-2,0^{ns}$	103
	cms-C	9,393		17,78		7,3		95
5.HST A483 - 2	normal	9,873	$+265^{ns}$	19,41	-0,69*	1,9	-0,1 ^{ns}	104
	cms-C	9,608		20,10		2,0		100
6.HST A483 - 33	normal	8,568	$+452^{ns}$	17,63	+0,06 ^{ns}	0,0	-0,1 ^{ns}	94
	cms-C	8,116		17,57		0,1		89
7.HST A483 - 39	normal	10,365	+578*	18,23	+0,72*	2,6	-2,3 ^{ns}	110
	cms-C	9,787		17,51		4,9		102
8.HST A483 - 38	normal	10,037	+310 ^{ns}	20,48	+1,19***	0,4	-1,1 ^{ns}	106
	cms-C	9,727		19,29		1,5		103
9.HST A483- 5	normal	10,006	+820***	17,27	0,00 ^{ns}	0,5	-0,1 ^{ns}	109
	cms-C	9,186		17,27		0,6		100
10.HST C344-427	normal	8,946	+210 ^{ns}	17,31	+0,59*	3,2	1,7 ^{ns}	95
	cms-C	8,736		16,72		1,5		95
Average - check	normal	9,709	+388***	17,93	+0,23*	3,0	-1,5 ^{ns}	103
	cms-C	9,321		17,70		4,5		97
Average trial		9515		17.82		3.75		100
LSD 0.05 $HxC=460$ $C_1-C_2=146$		6	HxC=0.59; C1-C2=0.19		HxC=4.4; C1-C2=1.4			
LSD 0,01	612	194		0.79	0.25	5.8	1.8	-
LSD 0,001	797	252	2	1.03	0.32	7.6	2.4	

Table 4. The influence of the two types of cytoplasm on some characters of the hybrids

*, **/ 00 , *** significant at 0.05, 0.01 and 0.001 probability levels, respectively; ns = non-significant; Nrf = normal cytoplasm.

CONCLUSIONS

The comparison of the grain yield of the hybrids produced based on male-sterile cytoplasm with normal analogues generally shows insignificant differences, except in 2022, when in conditions of water and heat stress, the hybrids obtained on normal cytoplasm showed more favorable behavior.

Only in the case of hybrids SURO 11, HST A483-5, and HST A483-39, the differences in grain yield between the two types of cytoplasm (normal and cms-C) were significantly higher in 2022, for isogenic fertile cytoplasm.

For the hybrids Turda 248, Turda 332, and Turda 2020, the yield differences between the hybrids produced on fertile cytoplasm and their male-sterile cytoplasm analogues were non-significant, in all three years of the field trials.

In the Turda 248 hybrid, the average grain yield obtained in the hybrid produced on

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male-sterile cytoplasm was higher compared to the yield of the fertile isogenic hybrid.

Hybrids: Turda 2020 (110-104%), Turda 332 (106-102%), HST A483-39 (110-102%), and HST A483-38 (106-103%) stood out for the highest selection index value (RSI), but also by the closest values between the two cytoplasm types on which the hybrids were produced.

The experimental data obtained show the ability to be used in the creation of hybrids and maize hybrid seed production of type male-sterile cytoplasm (cms-C), except for hybrids HST A483-5 and HST A483-39 in which the use of male-sterile cytoplasm significantly reduced grain production.

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.

The use of cytoplasm CMS-C to avoid manual detasseling is promising since it does not affect the agronomic performance of the hybrid.

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