CYTOPLASMIC DIVERSIFICATION EFFECT ON CERTAIN PLANT VEGETATIVE TRAITS OF SOME MAIZE (*ZEA MAYS* L.) INBRED LINES

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Abstract. *Isolines can be used for studies regarding the cytoplasmic influence of certain maize traits, for improving some of these traits but also for avoiding the cytoplasmic uniformity of the genetic basis. The aim of this paper was to identify any differences between five inbred line and their isogenic counterpart regarding the plant height, ear height, the number of leaves/ plant, leaf area and tassel length. The novelty of this study consists on the inbred lines used and the very specific conditions from the two experimental years. Several research were conducted on male sterile germplasm, but our study relate to fertile inbred lines. The biological material used consists of 25 inbred lines: 20 isolines and the five original inbreds. The isolines were obtained after transferring the nucleus of five inbred lines on four cytoplasm sources using the backcross method for ten generations. The inbred lines were studied in 2013 and 2014 at the maize breeding field of the Agricultural Research and Development Station Turda, Romania. There were identified two cytoplasm (T 248 and TC 221) that significantly influenced the plant and ear height, leaf area and the tassel length. These two cytoplasms can be used to improve these four traits. On this study we also found some interaction between the cytoplasm and the nucleus that caused significant differences when the isoline was compared to the original inbred, so each particular situation should be taken into consideration. The use of different cytoplasms caused increases or decreases for every trait; one cytoplasm can led to one trait increase for one inbred line, and decreases for another one. The results obtained can be used in further maize inbred line breeding programs, but also for obtaining more performant maize hybrids. The cytoplasms used in this study can be starting biological material for new isolines, but is very important to test their reaction to other inbred lines.*

Key words: *cytoplasm, isoline, maize*

INTRODUCTION

The studies regarding the cytoplasmic diversification were initiated after the discovery of the male sterility in maize and were fathom after the *Helmithosporium maydis* race *T* epidemic from the Corn Belt [1]. There were identified some ”per se” differences between inbred lines and the hybrids using these inbred lines when male sterile inbred lines were studied [2] [3] [4]. Some researchers found differences between an isoline and the original inbred line regarding the yield [3] [6] [7] [4] [8], cob traits [6] [9], plant vegetative traits [6] [10], biochemical composition [11] [12] [12] [9] [13], resistance to pests [2] and diseases [2] [15] [16].

Isolines can be used for studies regarding the cytoplasmic influence on certain maize traits, and for improving some of these traits. Another positive effect of creating isolines is the diversification of the genetic basis. A narrow genetic basis can lead to serious problems similar to southern corn leaf blight (*Helmithosporium maydis* race *T*) epidemic which caused important yield losses in the late 1960s in the USA.

At the Agricultural Research and Development Station Turda, in 1992, researchers from the maize breeding laboratory started an inbred lines improvement program by creating some isonuclear inbred line. The isolines were created by backcrossing cytoplasm donor inbred lines with the nucleus donor for ten generations. The lines were then mainteined by self-pollination. The program was also started for studies regarding the influence of these cytoplasms on the yield, cobs, plants, the biochemical composition, plant vigor, date of tasseling and silking, stalk lodging, the resistance to *Fusarium spp.* and *Ostrinia nubilalis*.

The objective of this paper is the study of the cytoplasmic effect on the plant height, ear height, number of leaves/ plants, leaf area and the tassel lenght on some maize inbred lines.

MATERIALS AND METHODS

The biological material consist of 25 lines: 20 isolines obtained after back-crossing four cytoplasm donor inbred lines and five nucleus donor lines; and the five original inbred lines. The nucleus donor inbred lines were used as recurrent parent in crosses that were realized for ten generations. The nuclei of TC 209, TC 316, TC 243, TB 367 and D 105 were transferred into four cytoplasm, coming from T 248, TB 329, TC 177 and TC 221. The transfer of the nuclei was realized in order to diversify the genetic basis of the inbred lines and to improve some traits. For obtaining a certain heterosis effect, the inbred lines used should belong to different heterotic groups, this aspect being take into consideration when the biological material was chosen. The cytoplasm and nucleus donor lines and their heterotic groups are presented in table 1.

Table 1.

Biological material- inbred lines heterotic group

	Inbred line	Heterotic group
Nucleus	TC 209	BSSS
	TC 316	Lancaster
	TC 243	BSSS
	TB 367	Argentinean Flint
	D 105	Fv 2 Flint
Cytoplasm	T 248	Lancaster
	TB 329	Iodent
	TC 177	Fv 2 Flint
	TC 221	European Flint

The inbred lines were studied in two experimental years (2013 and 2014) in the maize breeding field from the Agricultural Research and Development Station Turda, Romania, on a chernozem soil. The sowing density was 70000 plants/ ha, each plot consists of two rows: each 5 m long, 26 plants/ row. Each plot had five replications. Five plants from each plot were randomly picked for measurements, after pollination.

In 2013, the inbred lines were sowed in 29 April and harvested in 26 September, while in the second year the sowing was realized in 9 May and the harvesting in 14 October. In both years, the field technology was the same: 400 kg/ha NPK complex 27:13.5:0 from Azomures Romania, two manual hoeing and two herbicides were applied: preemergent 1.5 l/ha GoalTender (Dow AgroScience) and postemergent 2 l/ha Laudis 66 OD (Bayer CropScience).

The meteorological data was collected from Turda weather station (23° 47' E longitude; 46°35' N latitude), part of the Northern Transylvania regional Meteorological Centre.

RESULTS AND DISCUSSIONS

2013 and 2014 were very different regarding the temperature and precipitations during the maize growing season. 2013 had normal temperatures, with normal precipitation (compared to the 55 years average), but July was an arid month, with a rainfall deficiency of 39 mm. 2014 was a more favorable year for maize crop, due to its normal temperatures and heavy precipitations (67 mm above the 55 average in July and almost 28 mm in August).

Table 2.

Variance analysis for some plant traits of the studied inbred lines – Turda 2013-2014

Cause of variability	DF	Plant height (cm)	Ear height (cm)	No of leaves	Leaf area (cm^2)	Tassel lenght (cm)
		F test				
Total	249					
Years	1	44.5**	29.7**	1.2	162.3**	3.4
Repeats	4					
Nuclei	4	63.2**	85.7**	175.8**	77.7**	60.1**
Cytoplasms	4	12.4**	9.4**	1.1	5.7**	5.9**
Nuclei x Cytoplasms	16	6.2**	5.6**	3.4**	3.2**	2.6**
Years x Nuclei	4	15.4**	13.6**	17.6**	15.1**	1.4
Years x Cytoplasms	4	2.7*	4.4**	1.1	1.4	1.6
Years x Nuclei x Cytoplasms	16	1.8*	2.0*	2.7**	1.0	1.8*
Error (Y)	4					
Error (N)	32					
Error (C)	160					

The year significantly influenced the plant height, ear height and leaf area of the studied inbred lines, the values for the second year were higher that those from 2013. The nuclei and the interaction between nuclei x cytoplasm influenced in a significant way all studied traits. Acording to the variance analysis, except for the number of leaves, all trait were influenced by the cytoplasm, which led us to further study regarding the influence the cytoplasm have on these traits (table 2).

Table 3.

The cytoplasm influence on some maize plant traits of some inbred lines – Turda 2013-2014

Cytoplasms	Plant height (cm)	Ear height (cm)	No of leaves	Leaf area (cm^2)	Tassel lenght (cm)
	± original cytoplasm				
cyt T 248	3.5 **	3 ***	0.19	23.1 ***	0.97 **
cyt TB 329	-0.6	-0.4	0.13	10.1	0.11
cyt TC 177	-1.4	0.5	0.05	-1.6	-0.45
cyt TC 221	4.5 ***	3.8 ***	0.06	13.6 *	0.96 **
LSD 5%	2.1	1.7	0.20	11.9	0.7

LSD 1%	2.7	2.3	0.26	15.7	0.9
LSD 0.1 %	3.5	2.9	0.33	20.2	1.2

T 248 and TC 221 cytoplasms influenced in a significant way the plant height, ear height, leaf area and the panicle lenght (table 3). These cytoplasms could be therefore used for an improvement of the vegetative traits of the inbred lines. T 248 cytoplasm led to very significant increase of the leaf area of the plants: more that 23 cm^2. Some cytoplasms influenced the plant vegetative traits for several inbred lines, but some only in particular cases, due to the interaction between cytoplasm and nucleus, presented below.

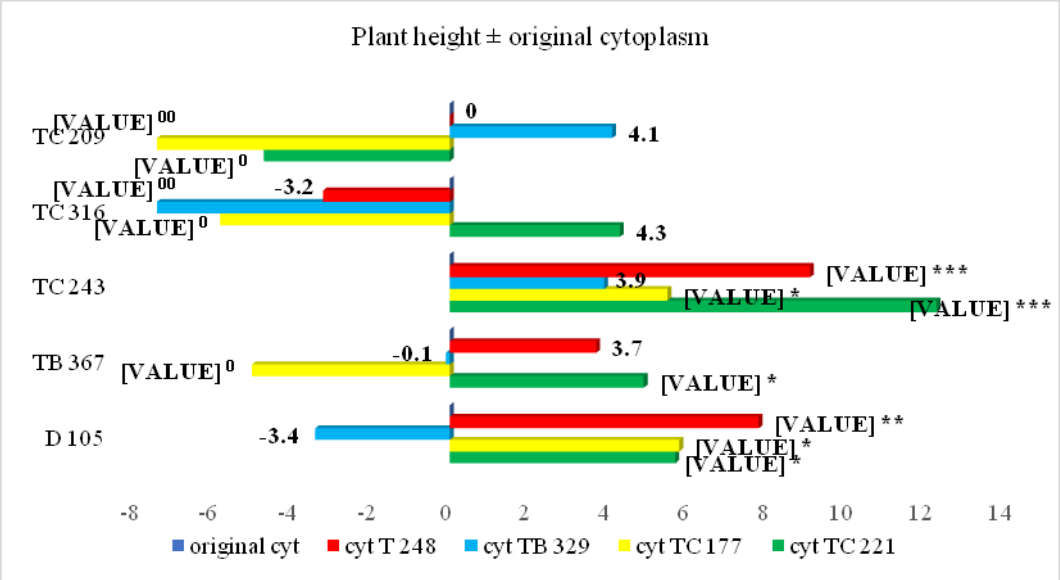

Figure 1. Cytoplasmatic effect on the plant height of some maize isolines – Turda 2013-2014

The use of T 248 and TC 221 cytoplasm on the five nucleus donor lines from our study led to significant increases of the plant height (table 3). It can be seen on figure 1 that these two cytoplasm did not have a significant influence for TC 209 and TC 316 inbred lines, but the height differences for the other three inbred lines were highly significant, for example + 12.3 cm for TC 243(cyt TC 221) compared to TC 243 inbred line. Using TC 177 cytoplasm caused significant plant height losses for TC 209, TC 316 and TB 367. TC 209 and TC 316 inbred lines have high values for the plant height (179 cm, respectively 185 cm), and this may be a cause of the negative effect of the cytoplasms. TC 243 has lower values for this trait (152 cm) so all cytoplasm increased it.

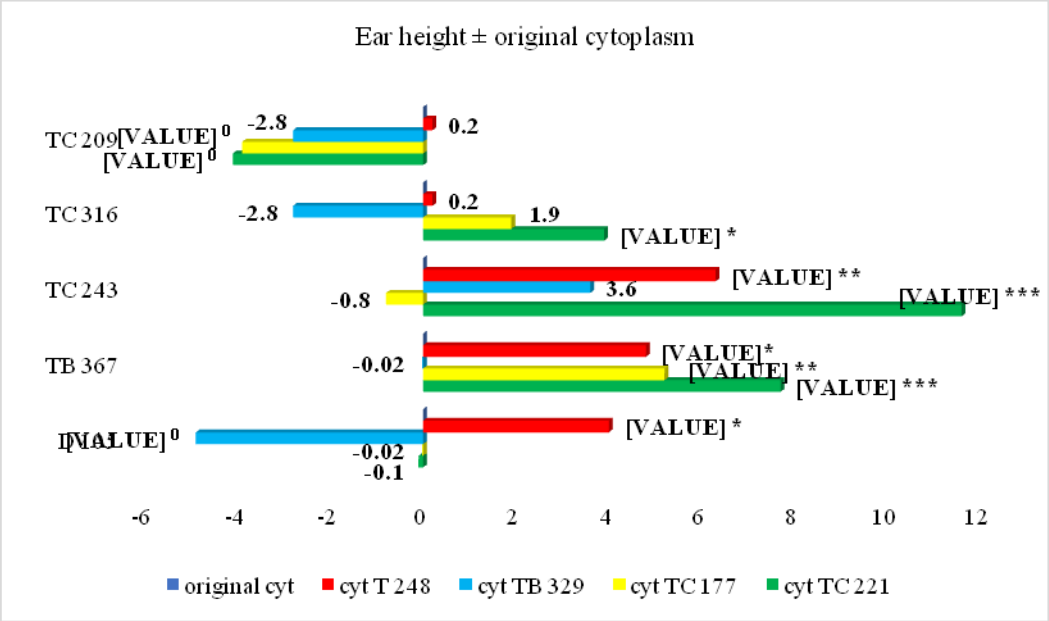

Figure 2. Cytoplasmatic effect on the ear height of some maize isolines – Turda 2013-2014

The two years ear height average of TC 209 was high (60 cm), so the interaction between this nucleus and the cytoplasms caused decreases for this trait. Using different cytoplasm sources increased the ear height for TC 316, TC 243 and TB 367. The largest difference was between TC 243 and its isogenic counterpart TC 243 (cyt TC 221), 11.6 cm. TC 243 had the lowest ear height, 36.6 cm and using T 248 and TC 221 cytoplasms can improve this trait. TC 221 had a negative influence on TC 209, a positive one on TC 316, TC 243 and TB 367 and no influence on D 105, so we can say that the effect of the cytoplasm is different depending on the cytoplasm x nucleus interaction (figure 2).

Using T 248 cytoplasm on TC 316, TC 243 and D 105 inbred lines, caused the increase of the leaf area in a significant way. Even if D 105 had the largest leaf area (440 cm^2), using T 248 and TB 329 cytoplasms caused significant increases for this trait; the largest difference between an isoline and the original inbred line was 67 cm^2 for D 105 (cyt TB 329). The leaf area of TC 316 was significantly influenced by T 248 and TC 221. Using TC 177 cytoplasm for TC 209 led to a decrease for this trait, this cytoplasm is the only one that caused smaller leaf areas. (table 4).

Table 4.

Cytoplasmatic effect on the leaf area of some maize isolines – Turda 2013-2014

Isoline		Original inbred line	± original
Isoline name	Leaf area	leaf area	cytoplasm
TC 316 (cyt T 248)	457	408	49 ***
TC 243 (cyt T 248)	386	341	45 *
D 105 (cyt T 248)	469	440	29 *
D 105 (cyt TB 329)	507	440	67 ***
TC 209 (cyt TC 177)	360	387	-27 0
TC 316 (cyt TC 221)	445	408	37 *
TC 243 (TC 221)	370	341	29 *
		LSD 5 %	26.7
		LSD 1 %	35.1
		LSD 0.1 %	45.2

The tassel lenght is an important trait, due to its negative correlation to the cob weight and the grain yield [2] [6]. In general, using different cytoplasms increased the tassel lenght, except TC 177 cytoplasm, which led in some cases to lenght decreases: 2.8 cm when used as cytoplasm for TC 316 (cyt TC 177) isoline. The tassel lenght of TC 316 was influenced in a significant way by three cytoplasms: T 248, TC 177 and TC 221, and the use of TB 329 and TC 221 influenced the tassel of D 105 (table 5).

Table 5.

Cytoplasmatic effect on the tassel lenght of some maize isolines – Turda 2013-2014

Isoline		Original inbred line tassel	± original
Isoline name	Tassel lenght	lenght	cytoplasm
TC 316 (cyt T 248)	41.7	40	1.7 *
TC 316 (cyt TC 177)	37.2	40	-2.8 *
TC 316 (cyt TC 221)	41.7	40	1.7 *
D 105 (cyt TB 329)	27.47	25.8	1.9 *
D 105(cyt TC 221)	27.8	25.8	2.0 *
		LSD 5 %	1.6
		LSD 1 %	2.1
		LSD 0.1 %	2.7

CONCLUSIONS

Two cytoplasms (T 248 and TC 221) significantly influenced the plant and ear height, leaf area and tassel length. There were also identified particular cases when the differences between the isoline and the original inbred line were caused by the cytoplasm x nucleus interaction. The two cytoplasms can be used to improve the plant vegetative traits, but also for improving certain inbred lines, the interactions between cytoplasm and nucleus should be taken into consideration.

The results obtained can be used in further maize inbred line breeding programs, but also for obtaining more performant maize hybrids. The cytoplasms used in this study can be starting biological material for new isolines, but is very important to test their reaction to other inbred lines.

The effect of the cytoplasm should also be tested in crossings with testers in order to study their influence on the simple hybrids.

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