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THE STUDY OF SOME INBRED LINES OF MAIZE AND HYBRIDS F1 RESULTED FROM THEIR CROSSING

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Abstract

The genetic diversity is an important source of heterosis, which is more intense when extremes types participate. Also the information on the genetic diversity of inbred lines is fundamental for the germplasm multiplication and conservation. In this paper, the characterization and establishment of the phylogenetic relationships between the Romanian and foreign inbred lines were performed by using the phenotypic differentiation index (PDI total) calculated for 13 characters and the heterosis. The results show that the phenotypic differentiation index (PDI) can be an effective tool in forecasting maize hybrids, but not always the greatest PDI means also the greatest heterosis. F1 hybrids resulting from Romanian crossed inbred lines extracted from the IInd selection cycle from the national germplasm collection, present both a good specific combining ability and a good productivity for the parental forms, being the best from an economic point of view in the given conditions.

Key words: *germplasm, heterosis, phenotypic differentiation index.*

INTRODUCTION

Maize (*Zea mays* L.) is one of the most valuable crops due to its multiple uses in food industry, animal husbandry and industry. The most important aspect concerning both researchers and farmers is the yielding capacity. The improvement of the yielding capacity begins with the selection of the inbred lines. The selection has to be associated with the testing of the general and specific combining ability as a measuring way of yielding capacity they give to hybrids.

Growing cultivated plant genetic resources, plant breeding and evaluation methods are important topics and they are also much debated. Present breeding programs aimed at searching older local populations valuable genes or other closely related species, including wild plants (Holubec, 2010).

Heterosis is a general biological character, which in the case of maize is both involved in morphological and physiological features. But the most important change in maize due to the heterosis bearing economic effects also that has led to the exploiting of this phenomenon by using hybrids between inbred lines is the reproductive heterosis (Has, 2004).

Today heterosis is considered to be an important means of increasing yield. The presence of enough hybrid strength is an important prerequisite for the successful production of hybrids and the heterotic studies can provide the basis for the exploitation of valuable hybrid combinations in the future breeding programmes (Thirupathi et al., 2012).

Thus, the largest maize growing countries (Romania included) cultivate this species mainly, growing especially simple hybrids (Voica, 2001). In Romania there are 2 721.2 thousand hectares (MADR, 2012) cropped with maize which makes it the first crop in order of cultivated surface.

The genetic diversity is very important when choosing inbred lines for hybridization programs (Banerjee and Kole, 2009).

There are many methods which have been used for the study of genetic diversity. The genetic diversity and its relationship with heterosis was studied by many authors (Hallauer et al., 1988; Smith, 1997; Bonea, 2001; Musat, 2008; Has et al., 2011).

Has et al. (2011) believe that the phenotypic evaluation of inbred lines is important since it prevents even many crosses, leading to a low cost within the breeding program and it also improves the assessment precision.
The estimation of the genetic variability of maize inbred lines based on phenotypic analysis is an important tool in order to carry out their identification and characterization. This type of analysis allows the observation of a species in relation to the plant's build and performances under biotic or a-biotic stress conditions since the environmental effect is always a stressful factor (Wietholter et al., 2008).
In Romania, in addition to the hybrids obtained from the local inbred lines (local populations and maize varieties cropped until the wide cropping of hybrids started to be generalized and used), foreign companies well known for their maize breeding and hybrid seed producing have introduced their own genetic material. This situation made us perform a comparative study of some Romanian and foreign inbred lines knowing that the information on the phenotypical and genetic diversity of inbred lines is fundamental for germplasm multiplication and conservation.

MATERIALS AND METHODS

The studied biological material was represented by ten inbred lines shown in Table 1, as well as six hybrids F1 resulted from crossing these lines.
The experiments took place at ARDS (Agricultural Research and Development Station) Simnic, Romania, in normal field conditions in 2009 and 2010 using the randomized block method with three repetitions. The ARDS is located at 44°19' N, 23°48' E, and 182 m height. The soils are luvosol.
Observations and grades have been made and given on the performance characters (grain yield, 1000 grain weight, grain-to-ear ratio, the note for the shriveled grain), plant's description elements (plant height, ear height, number of ears per plant, number of leaves per plant and foliar surface), ear characters (length, weight, diameter in the middle ear and the grade for the grain consistency).

Table 1. The studied biological material

Inbred lines and hybrids F1 resulted from crossing	The origin of inbred lines
Lc 235, Lc 151, Lc 64	Romanian inbred lines extracted from local populations from Oltenia, (obtained at ARDS Simnic)
Lc 235 x Lc 151	-
Lc 64 x Lc 235	-
Lc 408, Lc 406, Lc 403	Local maize germplasm extracted from the IInd selection cycle from national germplasm collection (obtained at National Agricultural Research and Development Institute -NARDI Fundulea)
Lc 408 x Lc 406	-
Lc 408 x Lc 403	-
Lc 9340, Lc 7445, Lc 4432, Lc 7619	Foreign inbred lines of German origin
Lc 9340 x Lc 7448	-
Lc 4432 x Lc 7619	-

For shrivelled of the grain there were given grades from 1 to 9 (1= very shriveled kernels; 9 = kernels full and well developed) and in the same way was done for the grain's consistency which received grades from 1 to 9 (1= dent, 9 = endured, glassy).
The genetic and phenotypic diversity of these ten inbred lines has been estimated based on:

- the calculation of the phenotypic differentiation index (PDI) using Herbert and Vincourt's formula (1985):

$$PDI = \sqrt{\sum (x_m - x_n)^2}$$

where: *Xm, Xn* – parental line average characters (m and n).

- in order to calculate the heterosis we used the formula proposed by Hallauer and Miranda (1981):

$$H\% = [F1-(P1+P2)/2]/[(P1+P2)/2]x100$$

where:

F1 = the average of the first hybrid generation and *(P1 + P2)/2* = average values for the parental forms P1 and P2.

RESULTS AND DISCUSSIONS

The average calculated values for the analyzed thirteen characters (four performance characters, five plant characters and four ear characters) cover the biometrical measurements on a two year period (2009 and 2010), when the experiments were carried out. These values express the *"per se"* value of a certain character for the inbred studied lines (Table 2). Making a comparison between the yield and its main elements for the studied inbred lines (Table 3) one can notice a superiority of the Romanian inbred lines. Higher grain yield for these Romanian inbred lines (10.0%) is mainly achieved due to ear weight (23.4%) and plants' height (10.4%).

The production capacity of inbred lines is an important aspect for the hybrid seed producer, but the real value of the inbred lines is given by their specific ability to carry through combinations. Therefore, the specific combining ability and the production capacity of Romanian and foreign maize inbred lines grown at ARDS Simnic have been intensively studied by Bonea and Urechean (2003), Bonea (2005), Bonea et al. (2009 a; 2009 b), Ilicevici (1994), Radu et al. (1994).

After the comparing (Table 4) the yield obtained and its main elements for the studied F1 hybrids we noticed a superiority of the foreign hybrids. The greater grain yield for the foreign hybrids (+13.5%) is achieved, especially, due to the higher vegetative strength showed by the foliar surface (+17.2%) and total plant height (+5.7%), but also due to the 1000 grain weight (7.9%).

The risks of vulnerability and the increase of genetic erosion for the *Zea mays* species have made necessary the search for new germplasm sources. As a result, for the breeder it is important to know the genetic differences between the used inbred lines, this being the only way how the crossings based on the heterotic model of the parental sources can be made successfully.

For an easier understanding of the phenotypic diversity of the studied inbred lines the differentiation indexes have been calculated for all the thirteen characters we analyzed (Table 5). Several authors (Herbert and Vincourt, 1985; Calboreanu – Badea, 2007; Musat, 2008; Copandean, 2010, 2011; Copandean and Rotar, 2012) have used phenotypic differentiation index in the study of the diversity of parental lines considering that the high value of this index shows a high degree of genotypic differentiation between the parental lines, while lower values indicate their similarity degree.

Analyzing the interaction between Romanian inbred lines extracted from the local populations one can notice that the lowest value, i.e. PDI = 31 (Lc 235 x Lc 151) and a middle value of PDI = 45 for Lc 64 x Lc 235 (Table 5). This means that between these inbred lines there is a small to medium genetic divergence since they come from a small geographical area.

At the interaction between Romanian inbred lines extracted from national germplasm collection one can notice that high values i.e., PDI = 82 (Lc 408 x Lc 406) and PDI = 88 (Lc 408 x Lc 403) are obtained (Table 5). There is a large genetic difference between these inbred lines meaning that they come from very different sources of germplasm.

Table 2. The performance characters and vegetative characters of the plant and ear for the studied inbred lines (the average at ARDS Simnic)

Characters	Inbred lines									
	Lc 235	Lc151	Lc 64	Lc 408	Lc 406	Lc 403	Lc9340	Lc 7445	Lc4432	Lc 7619
GY (t /ha)	5.70	6.00	6.85	7.02	5.64	5.83	5.76	5.10	5.14	6.21
TGW (g)	285	300	310	411	330	360	343	365	331	338
GER(%)	73	75	80	73	72	75	72	75	72	80
EP	1.14	1.13	1.15	1.14	1.01	1.00	1.00	1.04	0.99	1.15
PH(cm)	210	295	215	220	205	170	205	200	200	170
EH (cm)	70	80	83	80	50	52	60	67	75	60
SG*	8.5	8.0	8.0	8.5	8.0	8.5	7.5	6.5	8.5	8.5
LP^{-1}	12.8	13.0	15.6	13.0	13.1	12.1	16.0	13.7	15.3	13.2
FS (dm^2)	40.1	49.6	59.3	38.2	46.2	32.3	46.2	37.6	45.8	41.9
EL (cm)	16.3	14.9	18.4	18.0	17.4	14.1	16.9	18.0	16.0	17.4
EW (g)	167	190	200	171	167	198	179	138	148	170
ED (cm)	4.20	4.44	3.80	4.07	4.11	4.40	4.20	3.98	4.07	4.40
GC**	*2*	5	4	3	3	3	4	3	3	4

GY - Grain yield; TGW- 1000- grain weight; GER- Grain-to-ear ratio; EP- No. ears/plant; PH - Plant height; EH - Ear height; SG - Shrivelling of the grain; LP - No. leaves/plant; FS- Foliar surface; EL - Ear length; EW- Ear weight; ED - Ear diameter; GC - Grain consistency;

*1= very shriveled kernels; 10 = kernels full and well developed

**1= dent; 9 = endured, glassy

Table 3. The comparison of the yield and its main characters for the Romanian and foreign maize inbred lines

Characters	Romanian inbred lines average (control)	Foreign inbred lines average	Control difference	Control comparison %
GY (t/ha)	6.17	5.55	-0.62	-10.0
GER(%)	76.0	74.0	-2.0	-2.6
TGW (g)	332.6	344.2	+11.6	+3.5
EW (cm)	182.1	158.7	-23.4	-12.8
EP	1.09	1.04	-0.05	-4.5
PH (cm)	204.1	193.7	-10.4	-5.1
FS (dm^2)	44.2	42.8	-1.4	-3.1

GY - Grain yield; GER- Grain-to-ear ratio; TGW- 1000- grain weight ; EW- Ear weight; EP- No. ears/plant; PH - Plant height; FS- Foliar surface

Table 4. The comparison of the yield and its main characters for the Romanian and foreign maize hybrids

Characters	Romanian maize hybrids average (control)	Foreign maize hybrids average	Control difference	Control comparison %
GY (t/ha)	11.29	12.82	+1.53	+13.5
GER(%)	80.2	78.5	-1.7	-2.1
TGW (g)	438.0	473.0	+35.0	+7.9
EW (cm)	313.1	326.1	+13.0	+4.1
EP	1.03	1.05	+0.02	+1.9
PH (cm)	259.0	274.0	+15.0	+5.7
FS (dm^2)	67.8	79.5	+11.7	+17.2

GY - Grain yield; GER- Grain-to-ear ratio; TGW- 1000- grain weight ; EW- Ear weight; EP- No. ears/plant; PH - Plant height; FS- Foliar surface

Table 5. Phenotypic differentiation index of the maize inbred lines for 13 characters

Inbred lines	Inbred lines ♀				
♂	Lc 235	Lc 64	Lc 408	Lc 9340	Lc 4432
Lc 151	31				
Lc 235		45			
Lc 406			88		
Lc 403			82		
Lc 7448				48	
Lc 7619					42

Analyzing the interaction between foreign inbred lines we notice that we have obtained middle values for the PDI, from 43 (Lc 4432 x Lc 7619) to 48 (Lc 9340 x Lc 7448), which means that these lines originate from less genetically different sources.

Therefore, a lower differentiation index indicates a high degree of similarity between inbred lines. Similar results were obtained by Calboreanu - Badea (2007), Copandean and Rotar (2012).

The reproductive heterosis has represented and it still represents the main research goal on the heterosis at maize (Musat, 2008).

While in the first phase of introducing maize hybrids there have been obtained hybrids among varieties that gave a heterosis of 4.4 to 45.4% compared to the average of the parents, together with the resulted hybrids from the inbred lines, the intensity of reproductive heterosis has increased very much, especially due to the low productive capacity of the parental inbred lines (Has, 2004).

Table 6 presents the calculation of the value of the reproductive heterosis at the hybrids obtained from crossings between the studied inbred lines. For a more coherent data interpretation we split the hybrids obtained into three groups:

I. The hybrids obtained from Romanian inbred lines extracted from local populations in Oltenia (Lc 235 x Lc 151 and Lc 64 x Lc 235)

II. The hybrids obtained from Romanian inbred lines extracted from the IInd selection stage from the national germplasm collection (Lc 408 x Lc 406 and Lc 408 x Lc 403)

III. The hybrids obtained from foreign inbred lines (Lc 9340 x Lc 7448 and Lc 4432 x Lc 7619).

The reproductive heterosis obtained by the hybrids we studied, compared to the parents' average has been from 65.9 to 134.2%.

Analyzing comparatively the results on the intensity of heterosis one can notice differences between the three groups. The relative average value of the heterosis in the second and third group is close and for the hybrids in the first group is visibly lower (which can be due to a lower genetic difference between the inbred lines used for crossings, lines that have been obtained from local sources from a geographically small area, i.e. Oltenia).

Economically speaking, the best hybrids are the Romanian hybrids obtained at NARDI Fundulea (Lc 408 x Lc 406 and Lc 408 x Lc 403) whose parental lines have a good productivity.

Comparing the heterosis' intensity to the PDI (Table 5) values obtained by parental types of the hybrids we have studied one can see that the highest PDI does not always stand for the most clearly indicated heterosis. Similar results were obtained by Muşat (2008).

CONCLUSIONS

The phenotypic differentiation index (PDI) can be an effective tool in predicting maize hybrids, but the highest PDI does not always stand for the most clearly indicated heterosis.

The hybrids obtained from Romanian inbred lines extracted from local populations in Oltenia have a limited specific ability to carry through combinations, but they have a good productivity of parental lines.

The hybrids obtained from inbred lines extracted from the II[nd] selection stage from the national germplasm collection show both a good specific ability to carry through combinations and a good productivity of parental lines, being the most indicated economically speaking.

The foreign hybrids that we have studied have a very good specific ability to carry through combinations but a lower yielding capacity of parental lines under given conditions.

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