

## THE DIVERSITY OF SOME MAIZE INBRED LINES

Ana COPÂNDEAN, Carmen ROTAR

*Agricultural Research and Development Station  
Agriculturii str., no. 27, 401100, Turda, Cluj, Romania  
E-mail: copandean\_ana@yahoo.com*

**Abstract:** *Genotype diversity (morphological, ecological in the mechanisms of heterosis, through the action of the complementary favorable additive genes, favorable intrallelic and interallelic interactions, and even through the interactions between the nuclear genetic systems and the cytoplasmic factors. (MOLL et al. 1962, LAMKEY and EDWARDS 1999, ). The phenotypic and genetic diversity of maize inbred lines was particularly necessary in the creation of hybrids, being fundamental phenomenon of heterosis, the main route prevention of genetic vulnerability, thereby obtaining hybrid combinations preferred by growers or by seed producers. Results published by DUVIK (1999) that were compared hybrids and inbred lines of different selection periods converge to the conclusion that the extent of accumulation in advanced cycles of selection of favorable genes inbred lines of additive effects, production capacity „per se ” them is higher and heterosis values even if a lower capacity of the new hybrids is higher. By combining elements that help to achieve production capacity to reach a significantly higher heterosis. The study presents an estimate of the diversity of some new inbred lines, the heterosis and the relationship between diversity and heterosis. We used as parental forms 5 inbred lines considered as indicators of heterotic groups and 12 lines, new creations of the maize breeding team at S.C.D.A. Turda. By cross-breeding these lines, 70 simple hybrids were obtained that were experimented on in comparative cultures for 3 years. The obtained data was processed and we established the phenotypic differentiation index (IDF), the heterosis in the simple hybrids (Hallauer and Miranda 1981), the genetic diversity both at additive and non-additive levels, as well as the correlations between the diversity indicators. The results that we obtained lead us to the conclusion that the most efficient discrimination of the diversity of the inbred lines can be realized by simultaneously taking into consideration both quantitative indicators of diversity –  $Hr\%$ , IDF and  $ID\hat{g} + \Sigma s mn$ .*

**Key words:** *phenotypic, genetic diversity*

### INTRODUCTION

Plant amelioration, as an important branch of the conservation agriculture, makes heavy use of the genetic resources of vegetation and ads, apart from the genes productivity genetic material that improves the resistance to diseases, pests, and drought, with the purpose of creating organisms that are capable of lasting sustaining both agricultural production and environmental conditions.

Obtaining corn genotypes that are superior to the existent ones involves the development of new gene combinations and the number and value of these combinations depend largely on the diversity and the value of the collection that the breeder has available (SĂULESCU et. al. 2010).

The inbred lines of tropical and subtropical regions have a greater number of alleles and higher genetic diversity than those from the temperate zones. Inbred lines from temperate areas belonging to the "Stiff stalk Synthetic" (BSSS) heterotic group are on average the most divergent compared to all other inbred lines of the group (BIRCHLER 2003).

Some loci have relatively low levels of genetic variety, especially those loci that have been the subject of artificial selection, such as c1 and tb1 (GOODMAN, 2002).

The genetic value of maize inbred lines is determined by the source of germplasm that they are extracted from, the selection methods used during the successive inbreeding generations, as well as by the combination capacity expressed in the obtained heterosis.

In the field of applied genetics, quantifying genetic distance can be considered a tool for predicting heterosis, which serves to the prognostication of hybridization formulas with the best possible performance.

### RESEARCH OBJECTIVES

The priority objectives that need to be studied in the assessment of genetic diversity are numerous and need to be discussed systematically. Considering the importance and timeliness of research knowledge on phenotypic diversity and genetic inbred lines, our analysis methods pursued the following objectives:

- evaluating phenotypic and genetic diversity;
- controlling, conservation, utilizing and maintaining the existing germplasm on a rigorous scientific basis;
- comparing different methods for the assessment of diversity, with the intention of finding more effective ways of plant amelioration, than analyses based on molecular markers that can be considered relatively laborious and expensive.

### MATERIAL AND METHOD OF ANALYSIS

The biological material used in the conducted research was represented by: 5 inbred lines, considered to be indicators of the heterotic groups, and 12 lines, new creations of the maize breeding team at ARDS Turda. Estimating the diversity has been achieved by:

1. Calculating the phenotypic differentiation index according to Herbert and Vincourt 1985.

$$IDF = \sqrt{\sum (X_m - X_n)^2}$$

$X_m - X_n$  – average of parental line characters (m and n)

2. Calculating the heterosis according to Hallauer and Miranda 1981.

$$H\% = \frac{F_1 - \frac{P_1 + P_2}{2}}{\frac{P_1 + P_2}{2}} \times 100$$

H % – percentage expression of heterosis;

$F_1$  – the first hybrid generation;

$\frac{P_1 + P_2}{2}$  – average value of parental inbred lines  $P_1$  and  $P_2$  according to

Mackey, I., 1976

3. Calculating the genetic diversity:

- at the level of the homozygous loci for the additive genetic effects ( $\hat{g}$ )

- for the factorial system

$$\hat{g}_m \text{ or } \hat{g}_n = \frac{X_m}{m} \cdot \frac{X_n}{m.n} \quad (\text{Căbulea, 1975})$$

$X_m$  . – sum of the values in which the paternal parent participates constantly

$X_{..}$  – sum of the values in the factorial system

- at the inter and intra-allelic interactions, for the non-additive genetic effects ( $\hat{s}_{mn}$ )

- for the factorial system

$$\hat{s}_{mn} = X_{mn} - X_{..} - (\hat{g}_m + \hat{g}_n)$$

- for the diallel system

$$\hat{s}_{mn} = \frac{X_{mn} + X_{nm}}{2} - \frac{X_n - X_{mn} + X_n + X_n}{2} + \frac{X_n + X_n}{(p-1)(p-2)}$$

$\hat{s}_{mn}$  = genetic effect of the interactions of the genes of the two parents ( $X_m$  and  $X_n$ )

$X_{mn}$  or  $X_{nm}$  = phenotypic values of the hybrids

4. Calculating the correlations between the diversity indexes.

### RESULTS AND DISCUSSIONS

In tables 1 and 2 the lines that indicate the heterotic groups and the inbred lines created at Turda are shown, whose group heterotic is now known.

Table 1

Inbred Lines Indicators for Heterotic Groups

Inbred line	Origin	Genealogy	Heterotic group	Convariety variety	Heat units 50% silk
F 564	INRA Montpellier	F <sub>7</sub> x F <sub>64</sub>	<i>Euro-Late Flint</i>	<i>Indurata vulgata</i>	632
A 635	Minnesota	(ND203xB14)xB142	Stiff Stalk	<i>Dentiformis flavorubra</i>	672
A 619	Minnesota	(A171xOh43)xOh43	Lancaster	<i>Dentiformis xanthodon</i>	635
A 654	Minnesota	A116x WF <sub>9</sub>	Minnesota 13	<i>Dentiformis xanthodon</i>	587
W 153R	Wisconsin	Rec.I 153 R	I 153 R	<i>Dentiformis flavorubra</i>	632
MBS 847	-----	<i>Pioneer 3901</i>	Iodent	<i>Dentiformis flavorubra</i>	651

Table 2

Inbred Lines with Unknown Heterotic Group

Inbred line	Origin	Genealogy	Convariety variety	Heat units 50% silk
TC 335	S.C.A. Turda	(T248xT291)xTB329	<i>Dentiformis pyrodon</i>	622
TC 327	I.C.C.P.T.-Fundulea-S.C.A. Turda	(A632Euchlena)xP3780	<i>Aorista rubra</i>	614
TC 221	S.C.A. Turda	(Lo3 x Sint 1) x Lo3	<i>Indurata vulgata</i>	622
TC 344	S.C.A. Turda	GLG 480-139	<i>Dentiformis flavorubra</i>	636
TC 243	S.C.A. Turda	A654 x TC209	<i>Dentiformis xanthodon</i>	582
K 2308	Nord Saat Germania	MBS847 x D503	<i>Indurata vulgata</i>	592
TB 367	S.C.A. Turda	PI 187 x T248 I	<i>Indurata rubropaleata</i>	604
TC 365	S.C.A. Turda	TC208 x TB329	<i>Dentiformis flavorubra</i>	614
TC 331	S.C.A. Turda	(CO125 x B73) x A654	<i>Dentiformis flavorubra</i>	616
TC 314	S.C.A. Turda	T248 x TB329	<i>Dentiformis flavorubra</i>	625
TC 316	S.C.A. Turda	S54 x Mo17	<i>Dentiformis flavorubra</i>	609
TC 330	S.C.A. Turda	(CO125xB73)xTC209	<i>Indurata vulgata</i>	647

To characterize the differentiation at phenotypic level, the information provided by IDF was taken into account (table 3).

Analysis of the data in the table reveals the following:

- from F 564, the following lines are differentiated at an phenotypic level: TB 367 (IDF =97); K 2308 (IDF =85);
- from A 635, the following lines are differentiated: TC 243 (IDF=93); TB 367 (IDF=93); K 2308 (IDF 91); TC 316 (IDF= 82);
- from A 619, the following lines are differentiated: TB 367 (IDF =115); K 2308 (IDF)=99); TC327 (IDF= 74);
- from W 153, the following lines are differentiated: TB 367 (IDF =80); K 2308 (IDF=65); TC 316 (IDF=68); TC 344 (IDF =68); TC 344 (IDF =67); TC 335 (IDF =67);
- from MBS 847, the following lines are differentiated: TB 367 (IDF=92); K 2308

(IDF=84); TC 243 (IDF=79).

A lower differentiation index indicates a higher degree of similarity between the new lines and those that indicate the heterotic group.

Table 3

Phenotypic Differentiation Index of the Inbred Lines for 15 Traits

Line m n	F 564	A 635	A 619	W 153	MBS 847
TC 335	37	72	48	67	50
	0,998	0,995	0,997	0,994	0,997
TC 327	58	66	74	61	61
	0,997	0,998	0,994	0,995	0,998
TC 221	64	66	67	35	45
	0,994	0,996	0,994	0,996	0,998
TC 344	33	52	61	67	28
	0,998	0,998	0,995	0,996	0,999
TC 243	62	93	67	64	79
	0,998	0,998	0,908	0,997	0,998
K 2308	85	91	99	65	84
	0,994	0,998	0,991	0,997	0,997
TB 367	97	93	115	80	92
	0,991	0,997	0,985	0,993	0,994
TC 365	31	61	54	45	46
	0,999	0,991	0,997	0,997	0,999
TC 331	46	75	44	58	53
	0,997	0,995	0,997	0,995	0,997
TC 314	30	57	59	58	38
	0,998	0,997	0,995	0,995	0,987
TC 316	40	82	39	68	64
	0,998	0,950	0,998	0,997	0,999
TC 330	33	45	47	54	26
	0,991	0,998	0,997	0,997	0,999

Table 4

Expression of Reproductive and General Heterosis in Diallel and Cyclic Cross-Breeding

Line m n		F 564	A635	A 619	W 153 R	TB 329
TC 335	1	141	166	133	93	57
	2	94	103	92	70	62
TC 327	1	198	176	176	109	178
	2	117	104	102	80	104
TC 221	1	120	154	126	126	118
	2	87	98	88	89	83
TC 344	1	146	131	113	120	120
	2	96	90	81	88	82
TC 243	1	184	153	135	151	142
	2	111	99	91	99	93
K 2308	1	173	246	176	194	85
	2	107	136	111	116	69
TB 367	1	185	252	183	191	211
	2	111	135	115	116	119
TC 365	1	155	157	134	109	81
	2	101	113	92	84	74
TC 331	1	151	145	80	89	91
	2	96	93	71	77	72
TC 314	1	148	162	138	103	35
	2	98	103	94	86	54
TC 316	1	136	131	97	99	102
	2	91	92	94	86	77
TC 330	1	94	151	126	143	132
	2	77	54	99	104	95

- 1 % reproductive (the heterosis of grain production)
- 2 general heterosis =  $\frac{\% \text{ Reproductive} + \% \text{ Vegetative} + \% \text{ Development}}{3}$

% vegetative (the heterosis of the height of the plant, height of the cob insertion, total number of leaves, the number of leaves above the cob)

% development (100- % Heterozisul  $\Sigma t^0$  active at sowing – occurrence of stigmata)

The analysis of the diversity based on the intensity of expression of the heterosis (Hr%; Hg%) reflects a manifestation of heterosis with intensity levels rather differentiated (table 4). The reproductive heterosis (Hr%) was expressed through a very high amplitude, from 35% - 252%, thus highly discriminatory, while the amplitude of the heterosis (Hg%) was characterized by lower levels 54% - 135%. Among the most intense expression of heterosis, we can mention those of the line: K 2308 with A 635 (Hr%=246; Hg%=135); K 2308 with W153 (Hr%=194; Hg % =116); TC327 with F 564 (Hr%=198; Hg%=117); TC243 with F 564 (Hr%=184; Hg%= 111); TB 367 with F 564 (Hr%=185; Hg %=111); K 2308 with A 619 (Hr%=176; Hg%=111) and W 153 R (Hr% =194; Hg %=116); TB 367 with MBS 847 (Hr%=211; Hg %=119).

A lower intensity heterosis indicates a possible genetic relationship among the lines: TC335 with MBS 847, as well as between the lines: TC 314 with MBS 847, otherwise confirmed by the phenotypic differentiation index presented in table 3.

The correlations between the IDF specific to different characters of the different indicator lines and the corresponding heterosis are shown in table 5. The most discriminatory character can be considered the grain production ( $r=0.498^*$ ) and the most discriminatory line can be considered line A 619 ( $r=0.679^*$ ).

Table 5

Correlations Between Phenotypic Diversity of Parental Forms (IDF) and the Character-Specific Heterosis (H%)

Heterosis	From the Indicator Line					
	F 564	A 635	A 619	W 153 R	MBS 847	General
Grain production	0,495*	0,516*	0,735**	0,299	0,315	0,498**
Number of rows	0,446	0,043	0,462	0,084	0,030	0,220*
Number of grain/row	0,406	0,213	0,699**	0,207	0,150	0,391**
Length of the cob	0,562*	0,343	0,655**	0,134	0,267	0,436**
Depth of the grain	0,628*	0,413	0,686**	0,296	0,457	0,516**
$\Sigma t^0$ sowing - occurrence of stigmata	-0,338	0,338	-0,143	0,240	0,118	0,126
General	0,491*	0,579*	0,679**	0,327	0,283	0,495**
r pt P 5% (GL15)			0,482			0,217(GL8)
1% (GL15)			0,606			0,283(GL8)

Genetic differentiation at an additive level was achieved by calculating the genetic differentiation index ( $ID\hat{g}$ ) and/or the correlation between the additive effects ( $r\hat{g}$ ). Analyzing the data from table 6, we found a relative concordance between the estimates based on ( $ID\hat{g}$ ) and ( $r\hat{g}$ ) especially with the statistical limitations. The new lines had a different behavior from the indicator lines:

- from line F 564, the following lines are genetically differentiated at an additive level: TC 327; K 2308; TC314;

- from line A 635, the genetic differentiation was observed especially in line TC 243;
- from line A 619, we can consider differences for lines TC 335 and TB 367;
- from line W 153, these lines are different: TC 344 and TC 365;
- from line MBS 847, lines TC 331, TC316 and TC330 are different.

*Table 6*

Genetic Diversity of New Inbred Lines at the Level of Additive Loci (ID $\hat{g}$ , r $\hat{g}$ ). m x n Crossing System

Line		F 564	A 635	A 619	W 153 R	MBS 847
TC 335	1	22.43	6.98	29.63	20.45	11.11
	2	-0.2916	0.8431*	-0.6496*	-0.3037	0.5262*
TC 327	1	36.60	32.65	20.31	21.83	31.27
	2	-0.6236*	0.3880	0.4941*	0.3726	-0.0483
TC 221	1	25.41	13.94	32.82	22.45	28.32
	2	-0.1110	0.6663*	-0.3736	0.1026	-0.0010
TC 344	1	19.06	17.10	36.12	33.04	34.17
	2	0.5304*	0.6301*	-0.2368	-0.6284*	-0.1702
TC 243	1	22.50	28.01	19.94	24.75	27.72
	2	0.2636	-0.6127*	0.4140	-0.3294	-0.0430
K 2308	1	60.21	45.78	44.52	30.62	35.58
	2	-0.5312*	-0.3475	-0.2520	0.7227*	0.3998
TB 367	1	32.92	28.59	36.96	18.53	22.00
	2	-0.3423	-0.1875	-0.5449*	0.6132*	0.4948*
TC 365	1	9.81	16.01	28.75	23.77	23.44
	2	0.7765*	0.0598	-0.4055	-0.6793*	0.1377
TC331	1	11.29	19.90	19.57	18.71	29.41
	2	0.7273*	-0.4314	0.4072	-0.1336	-0.6108*
TC 314	1	31.14	19.55	31.65	27.20	11.67
	2	-0.5539*	0.3246	-0.3434	-0.4657	0.8345*
TC 316	1	17.64	30.29	24.92	32.83	43.22
	2	0.7636*	-0.1983	0.5192*	-0.3617	-0.7262*
TC 330	1	23.78	22.00	24.31	24.31	38.43
	2	0.1231	0.0908	-0.0531	-0.0531	-0.8199*

1 – ID $\hat{G}$

2 – r $\hat{g}$

\* significant differences (-r) or similarities (+r)

Intra and interallelic gene interactions are shown quantitatively through the effects of the specific combination capacity ( $\hat{S}_{mn}$ ), in table 7.

Positive values show a higher degree of differentiation of the interactive genes; negative values show the degree of similarity.

- from line F 564, the following lines are differentiated on a genetic non-additive level: TC 327 and TC 365;
- from line A 635 we can consider as genetically differentiated at non-additive level the lines TC335; K 2308; TB 367; TC 314 and TC 316;
- from line A 619, the following lines are relatively genetically differentiated at non-additive level: TC 335; TC314 and TC 365;
- from line W 153 R, the following lines are genetically differentiated at non-additive level: TB 367 and TC 316;
- from MBS 847, the lines that have genetic differences at non-additive level are: TC 327; TC 221; TC 243 and TC 316.

Table 8 presents an analysis of the correlation between  $ID\hat{g}$  and heterosis. We observed a significant correlation ( $r=0.266^*$ ) only with the expression of the reproductive heterosis.

Between  $ID\hat{g}$  and  $H\hat{g}$  there was no significant correlation ( $r=0.025$ ), which suggests that the reproductive heterosis could be considered as a more accurate expression of the diversity of homozygous loci than of the general heterosis.

The analysis of the correlation between the heterosis and the intra and interallelic interactions shows the lack of correlation between the two variables ( $r=0.157$ ;  $0.136$ ).

Analysis of the correlation between the additive effects and the non-additive interactions with the reproductive and general heterosis shows the significantly positive correlation ( $r = 0.380^*$  and  $r = 0,260^*$ ).

*Table 7*

Genetic Diversity of New Inbred Lines vs. Indicator Lines Based on Non-Additive Effects ( $\Sigma \hat{S}_{mn}$ )  
m x n Crossing System

Lines m n	F 564	A635	A619	W153R	MBS 847
TC 335	7.09	38.18	61.95	-44.95	-62.29
TC 327	59.63	-67.89	-42.62	1.10	49.78
TC 221	-16.26	-16.57	0.39	6.31	26.21
TC 344	8.14	-28.08	-4.71	17.68	7.03
TC 243	19.11	-18.09	-45.95	7.50	37.45
K 2308	-40.82	22.32	20.85	21.15	-20.46
TB 367	-72.11	33.79	-20.11	36.94	-25.41
TC 365	24.98	-3.74	38.63	-34.44	11.83
TC 331	16.41	4.63	-21.97	-16.94	-87.88
TC 314	0.75	26.03	61.98	-9.56	-3.61
TC 316	14.82	27.81	-51.33	32.06	53.47
TC 330	-38.24	-14.85	-3.74	3.74	22.51

*Table 8*

Correlations between the Genetic Diversity Indicators and Heterosis

Indicators	Hr %	Hg %
$ID\hat{g}$	0,266*	0,025
$r\hat{g}$	-0,014	-0,051
$\Sigma \hat{S}_{mn}$	0,157	0,136
$ID\hat{g}$ și $\Sigma \hat{S}_{mn}$	0,380**	0,260*

\* 0,250 significant for P 5%

\*\* 0,325 significant for P 1%

### CONCLUSIONS

1. Differentiating genotypes based on IDF proved to be more discriminant than by using the correlation coefficient between the phenotypic expression of the characters ( $r_f$ ).

2. The heterosis of the phenotypic characters has manifested in a way specific to each character and each experimental condition; the most representative and discriminant being the one calculated based on the multi-annual averages and expressed by the reproductive heterosis ( $H_r\%$ ).

3. There is a similarity in expressing the relationship between the reproductive and general IDF and  $H\%$ , therefore utilizing a single analysis should be sufficient in order to

classify the lines.

4. Calculating the additive effects corresponding to the inbred lines allowed us to estimate the improvement value of the lines, the lines that carry genes suitable for production, elements of production, as well as the resistance to braking and falling (TC 344; TC 365; TC 243).

#### **BIBLIOGRAPHY**

1. BIRCHLER J A 2003. Genetic Structure and Diversity Among Maize Inbred Lines as Inferred From DNA Microsatellites Genetics, Vol.165, 2117-2128.
2. BLEY, J.F., GOODMAN, M. M., STUBER, C.W., 1986. Exceptional genetic divergence of Northern flint corn. American Journal Botany, 73:64-69.
3. GOODMAN, M. M. 2002. Genetic diversity and selection in maize starch pathway.
4. HEBERT, Y., VINCOURT, P., 1985 Mesures de la divergence genetique Distances Calculees sur des criteres biometriques. Estimation et applications INRA Station d'Amelioration des Plantes fourrageres Lusignan.
5. LAMKEY, K. R. 1992. Fifty years of recurrent selection in the Iowa stiff stalk synthetic maize population Maydica 37:19-28.
6. MOLL, R.H 1962. Heterosis and diversity in variety crosses of maize Crop Sci. vol 2 nr.3 197-198.
7. SĂULESCU, N., ITTU G., GIURA, A., MATILDA CIUCA, MUSTĂŢEA P., MARIANA ITTU., GABRIELA FĂNTĂNĂ, FLORENTINA AMALIA NEACŞU 2010. Diversitatea bazei genetice ca fundament al progresului în ameliorarea grâului. Anale INCDA Fundulea, VOL .LXXVIII Nr.1
8. SMITH, J.S.C., 1988 Diversity of United States hybrid maize germoplasm; isozymic and chromatographic evidence Crop Sci. 28:63 – 69.