

## DETERMINING THE DEGREE OF RELATEDNESS / DIFFERENTIATION BETWEEN SOME MAIZE INBRED LINES OBTAINED FROM TWO SYNTHETIC TU SRR COMP. A AND TU SRR COMP. B

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**Abstract.** Differentiation of maize inbred lines in order to assess their degree of relationship and their classification into germplasm groups or in heterotic groups is performed using different methods. For the primary differentiation the method of pedigree is used, which gives us information on the origin of the initial material of selection. Knowledge of the relationship between diversity and the maize inbreds has a significant impact on establishing maize breeding programs in creating hybrids for framing inbred lines germplasm in groups and for creating original biological material. The paper aim is to determine the degree of relatedness/ differentiation of the lines created from two synthetics (Tu SRR Comp. A and Tu SRR Comp. B). The two synthetic composites were created starting from the late maturity inbred lines B 73 (Tu SRR Comp.A) and Mo 17 and C 103 (Tu SRR Comp. B). B 73 inbred line is part of the Stiff Stalk Synthetic germplasm, while Mo 17 and C 103 belong in the Lancaster Sure Crop group. For Tu SRR Comp A there were used as parental genotypes A632, CM 105, TB 329 and T 291 inbred lines, while for the second synthetic composite, besides the two inbred lines from the Lancaster Sure Crop, there were also used T 248, W633 and TC 208. The biological material used consisted of eight inbred lines and 28 hybrids. Four inbred lines were obtained from Tu SRR Comp. A and four from Tu SRR Comp. B. between these inbred lines, there were performed (4)  $p(p-1)/2$  diallel crossing, resulting in 28 hybrids. The study was conducted in the two experimental years: 2013 and 2014, at the Agricultural Research and Development Station Turda. The experimental model used was randomized block with four repeats. There were analyzed an important number of traits: for inbred lines: plant height, ear height, leaf area, total number of leaves/plant, number of leaves above the ear, number of tassel ramifications, tassel length and tassel main axis, while for hybrids there were taken into consideration the plant height, ear height, leaf area and the total number of leaves/plant. In order to determine the factors involved in the expression of the traits we used the analysis of variance and the orthogonal variance decomposition into its components, inbred lines pedigree. There were performed biometric measurements of the phenotypic characters and general combining ability was calculated.

### INTRODUCTION

The history of the two composite populations Tu SRR Comp. A (Comp. B) and Tu SRR Comp. B (Comp. A) began in 1985 when it was decided to constitute two composites based on the heterotic model B73 x Mo17 (Haş I., personal communication).

The hybrid combination B73 x Mo17 and its variants (earlier or later) occupied in the late twentieth more than 60% of the maize from the Corn Belt of the United States of America (TROYER, 1999, HADI, 2004. HALLAUER, 2003. HAŞ I., 2004. SARCA, 2004, HALLAUER ET AL. 2010).

Composites can originate also from inbred lines, the number of inbred lines being less than eight (HAŞ I., 2004). The two synthetic composites were created starting from the late maturity inbred lines B 73 (Tu SRR Comp.A) and Mo 17 and C 103 (Tu SRR Comp. B). B 73 inbred line is part of the Stiff Stalk Synthetic germplasm, while Mo 17 and C 103 belong in the Lancaster Sure Crop group. For Comp. A there were also used as parental genitors A632, CM105, TB329 and T291, and for Comp.B in addition to the two Lancaster Sure Crop inbred

lines were used as the parental form T 248, W 633 and TC 208. Crosses were done in 1985, and the inbred lines B73 respectively C103 and Mo17 were used as maternal forms. In 1986 there were created the double hybrids. Hybridization were conducted so that the Comp. A inbred line B 73 to have 50% share and in Comp. B, Mo17 and C103 to have 50% share.

In 1987, separately for each of the two composite populations there were used combinations of double hybrids detained and in the field there were sowed from the balanced hybrid mixture (to be pollen supplier) and double hybrids to be use as maternal form (HALLAUER AND CARENA, 2009). Heterotic groups are used due to Falconer (1960) demonstration that expression of heterosis is dependent on the non-additive gene effects, and the magnitude of heterosis is dependent on the difference between the frequency of the allele.

It was considered that the two composite populations will simultaneously meet two requirements:

- earlier vegetative period for some valuable germplasm sources as the heterotic model B73 x Mo17 (FAO-650-700) was totally unsuitable for our area with limited thermal resources;
- maintain the heterosis between two inbred lines derived from the two heterotic groups.

It is necessary that complementary heterotic groups to have in their composition germplasm that, by crossing the inbred lines resulted to produce hybrids that can outperform those from crossings between inbred lines from the same heterotic group. Evolution of heterotic groups should be considered by the accumulation of secured favorable genes (additive effects) in both composite populations and to realize through crosses, the non-additive effects (HAS, 2004). Currently used as a reference for this type of selection and improvement of inbred lines, the initial materials from BSSS and non-BSSS heterotic groups (MIKEL AND DUDLEY, 2006).

To establish relations between various sources of germplasm and genetic diversity of inbred lines there were used various methods:

- **pedigree method** provides genealogical information on the origin of original selection material (Smith, 1988; Smith et al., 1990);
- **biochemical and molecular technologies** based on DNA fingerprinting were shown to be useful in studies of genetic similarity on the basis of PCR, RAPDs, SSR și AFLPs (Smith and Smith, 1987; Melchinger, 1993; Smith et al., 1997; Pajic, 1998; Senior et al., 1998; Şuteu Dana et al., 2013).

The objective of this study was to evaluate the relationship of relatedness and / or genetic diversity between eight inbred lines created from ARDS Turda, using specific methods:

- phenotypic methods based on the analysis of the biometrics;
- genetic methods based on: - pedigree analysis;
- correlations of the genetic parameters

## **MATERIAL AND METHODS**

For the achievement of the objective there were used eight inbred lines, 4 inbred lines obtained from Tu SRR Comp. A (Comp. B) belonging to the BSSS germplasm group and 4 lines derived from Tu SRR Comp. B (Comp. A) from the Lancaster germplasm group.

Table 1

Inbred lines origin and genealogy			
Current number	Inbred line	Origin	Genealogy
<b>Inbred lines BSSS group</b>			
1.	TD 337	Sel. Turda Comp.A	H-8-1-2
2.	TA 426	Sel. Turda Comp.A	H20-2-4-
3.	TA 428	Sel. Turda Comp.A	H33-1-2-
4.	TA 422	Sel. Turda Comp.A	6088-1-1
<b>Inbred lines Lancaster group</b>			
5.	TC 385 A	Sel. Turda Comp.B	H60-1-1-
6.	TC 384 A	Sel. Turda Comp.B	H46A-1-2-
7.	TC 398	Sel. Turda Comp.B	H77-6-7-
8.	TC 399	Sel. Turda Comp.B	H84-6-7-

The biological material used is the F1 hybrids realized in (4) p (p-1) / 2 type diallel crosses (CABULEA, 1975), meaning 8 inbred lines and the resulting 28 hybrids. The inbred lines and the hybrids were experienced in 2 years and 4 repetitions.

Field experimentation of the hybrids and their parental genotypes was conducted at the Agricultural Research and Development Station Turda in 2013 and 2014 on plots of 7.0 m<sup>2</sup> on randomized blocks with four repetitions at a density of 60,000 plants/ ha.

Climatic condition between May and September during the two years of experimentation (2013-2014) were different. 2014 was a more favorable year for maize crop, following a surplus of rainfall and normal temperatures in June, July and August. 2013 was normal in terms of precipitations during the maize growing season, but noting that in July there was a shortage of precipitations of 39.1 mm that influenced pollination and grain yield per hectare. In terms of temperature, we can say that 2013 was a warm year throughout the growing season of maize.

In order to determine the factors involved in the expression of traits were used the analysis of variance and the orthogonal decomposition of variants components variance.

The variance of hybrids has been decomposed into:

- variance of hybrids with parental forms from Tu SRR Comp. A (Comp.B)
- variance of hybrids with parental forms from Tu SRR Comp. B (Comp.A)
- variance of hybrids with parental forms from Tu SRR Comp. A (Comp. B) x Tu SRR Comp. B (Comp.A)
- comparison between hybrid groups.

## RESULTS AND DISCUSSIONS

In (Table 1), in which is presented the analyze the genealogy for the 8 inbred lines studied, it can be observed that although they have the same origin (pedigree) they belong to different families, families which have diverged during the stabilization process through selection for specific phenotypic traits.

By the analysis of phenotypic characters (Table 2) it can be seen significant differences between the two inbred lines groups in terms of: plant height (207 to 179), ear height (69 to 56), total number of leaves/plant (13.8 to 12.6), tassel main axis lenght (21.4 to 22.9), tassel length, number of tassel ramifications, number of leaves above the ear. Between the lines obtained from Tu Comp.A (Comp. B) there is greater variability that between the inbred lines whose starting material was Tu SRR Comp. B (Comp. A) for the three characters; therefore we can highlight the existence of heterogeneity of the composit Tu SRR Comp. A (Comp. B).

Table 2

Some *per se* traits of the eight studied inbred lines

Inbred line name	Plant height		Ear height		Tassel main axis length		Tassel length		Number of tassel ramifications		Total number of leaves/plant		Number of leaves above the ear	
	cm	± M1	cm	± M1	cm	± M1	cm	± M1	no	± M2	no	± M2	no	± M2
TD 337	21.5	8*	81	12**	17.4	-4 <sup>000</sup>	25.5	2.9 <sup>00</sup>	12.7	2.7**	13.8	0	5.4	0.4 <sup>00</sup>
TA 426	20.3	-4	66	-3	21.7	0.3	28.4	0	12.5	2.5**	14	0.2	6	0.2
TA 428	19.8	-9 <sup>0</sup>	64	-5 <sup>0</sup>	21.2	-0.2	27.4	-1	10	0	13.5	-0.3	5.6	-0.2
TA 422	21.3	6	67	-2	25.3	3.9**	32.1	3.7**	6.5	-3.5 <sup>00</sup>	13.9	0.1	6.1	0.3*
<b>Mean -M1</b>	<b>207</b>		<b>69</b>		<b>21.4</b>		<b>28.4</b>		<b>10</b>		<b>13.8</b>		<b>5.8</b>	
	cm	± M2	cm	± M2	cm	± M2	cm	± M2		± M2		± M2		± M2
TC 385A	18.1	2	41	-15 <sup>00</sup>	20.3	2.6**	27.8	-1.1	12	1	12.2	-0.4	6.1	0.8**
TC 384A	16.6	-13 <sup>00</sup>	59	3	25.9	3***	30.3	1.4	12	1	12.4	-0.2	4.3	-1 <sup>000</sup>
TC 398	16.0	-19 <sup>00</sup>	49	-7 <sup>00</sup>	21.0	-1.9 <sup>00</sup>	26.7	-2.2 <sup>00</sup>	11	0	11.6	-1 <sup>00</sup>	5.8	0.5**
TC 399	20.8	29**	73	17**	24.3	1.4*	30.8	1.9**	10	-1 <sup>000</sup>	14.1	1.5**	5.1	-0.2
<b>Mean -M2</b>	<b>179</b>		<b>56</b>		<b>22.9</b>		<b>28.9</b>		<b>11</b>		<b>12.6</b>		<b>5.30</b>	
<b>M1 - M2</b>	<b>28.3</b>		<b>13.3</b>		<b>-1.5</b>		<b>-0.5</b>		<b>-1</b>		<b>1.2</b>		<b>0.50</b>	
Significance	***		***		000		000		000		***		***	
LSD (P5%)	6.81		4.15		1.16		1.41		1.30		0.63		0.25	
LSD (P1%)	9.11		5.55		1.55		1.89		1.74		0.84		0.33	
LSD (0.1%)	11.96		7.29		2.03		2.47		2.28		1.10		0.44	

Table 3

Analysis of variances for plant height and insertion height cob averages of the hybrids grouped by the origin of their parental inbred lines, ARDS Turda 2013-2014

Source of variability	DF	Plant height		Ear height	
		cm		cm	
		Mean	s <sup>2</sup>	Mean	s <sup>2</sup>
Total	223				
Years (y)	1		65195**		6418**
Repetitions	3				
Error (A)	3		270		43.5
Genotypes (G)	27		2112**		976**

1. Hybrids between A inbred lines x A inbred lines (AxA) <sup>a</sup>	(5)	248	565**	83.8	221**
2. Hybrids between B inbred lines x B inbred lines (BxB) <sup>b</sup>	(5)	222	1629**	68.7	745**
3. Hybrids between A inbred lines x B inbred lines (AxB)	(15)	248	430**	82.2	277**
Comparison between hybrid groups: (AxA), (BxB), (AxB)	(2)				
Comparison between hybrids: (AxA) and (BxB)		25.67***		15.04***	
(AxA) and (AxB)		-0.32		1.58	
(BxB) and (AxB)		-25.99 <sup>000</sup>		-13.46 <sup>000</sup>	
Genotypes x Years	27		93.61*		44.86
Error (G)	162		61.65		34.56
LSD (P5%)		7.77		5.82	
LSD (P1%)		10.24		7.67	
LSD (P 0.1%)		13.19		9.87	

The analysis of variance for plant height and ear height (Table 3) reveals the importance of the years for hybrids expressing, both for those resulting from crossing inbred lines inside the group and those between other tested groups. Through decomposition of orthogonal it was possible to highlight the significant differences both between hybrids resulted from crossings between inbred lines from the same group and of hybrids resulted after crossings between different groups. There are noted very positive significant differences for plant height between [(AxA) - (BxB)] hybrids and very significant negative differences between [(BxB) - (AxB)]. For the ear height there are very significant positive differences even between hybrids obtained by crossing inbred lines of the same group [(AxA) - (BxB)] and significant negative differences for the crossings inside the [(BxB) - (AxB)] group.

The analysis of variance for the total number of leaves and leaf area (Table 4) shows that there are significant differences, for both traits within each of the groups of hybrids compared. And for these characters the experimental years influence and of the interactions between years and hybrids were distinctly significant.

Table 4

Analysis of variances for total number of leaves/plant and leaf area averages of the hybrids grouped by the origin of their parental inbred lines, ARDS Turda 2013-2014

Source of variability	DF	Total number of leaves/plant		Leaf area	
				cm	
		Mean	s <sup>2</sup>	Mean	s <sup>2</sup>
Total	223				
Years (y)	1		103**		388994**
Repetitions	3				
Error (A)	3		0.97		8013
Genotypes (G)	27		4.16**		12692**
1. Hybrids between A inbred lines x A inbred lines (AxA) <sup>a</sup>	(5)	13.03	3.42**	500	77089**
2. Hybrids between B inbred lines x B inbred lines (BxB) <sup>b</sup>	(5)	13.07	8.17**	533	16307**
3. Hybrids between A inbred lines x B inbred lines (AxB)	(15)	13.59	49.23**	554	30291**
Comparison between hybrid groups: (AxA),	(2)				

(BxB), (AxB)					
Comparison between hybrids:					
(AxA) cu (BxB)		-0.04		-32.71	
(AxA) cu (AxB)		-0.56 <sup>0</sup>		-53.47 <sup>00</sup>	
(BxB) cu (AxB)		-0.52		-20.76	
Genotypes x Years	27		2.35**		2145**
Error (G)	162		44.54		1095
LSD (P5%)		0.52		32.75	
LSD (P1%)		0.68		43.18	
LSD (P 0.1%)		0.88		55.59	

The analysis of genetic diversity was carried out at the additive gene action level, respectively at the overall gene effect (G) characteristic to the eight inbred lines (Table 5).

Table 5

Additive genetic effects (G) corresponding parental inbred lines Turda, 2013-2014

Traits	TD337	TA426	TA428	TA422	TC385A	TC384A	TC398	TC399
Plant height	7.9	7.1	4.7	2.4	-1.4	-10.6	-17.1	-7.0
Ear height	6.7	5.6	0.4	0.3	-9.4	-2.3	-9.3	8.0
Tassel length	-0.7	-0.5	0.1	0.8	0.2	0.6	-0.5	0.1
Tassel main axis length	-0.7	-0.8	-0.6	1.4	-0.1	0.8	-0.6	0.5
Number of tassel ramifications	1.3	1.1	0.3	-1.5	-0.3	-0.1	1.3	-2.1
Total number of leaves/plant	0.0	0.3	0.0	-0.3	-0.1	-0.5	-0.4	0.3
Number of leaves above the ear	-0.1	0.2	-0.2	0.1	0.4	-0.5	0.2	-0.1
Leaf area	8.9	1.3	-2.9	-35.9	8.1	14.3	-7.3	13.4

\_lines that remark very significant by their capacity to transmit the analyzed traits

From the analysis of combining ability of the eight inbred lines some distinguished by the ability to transmit several traits. Some valuable inbred lines regarding this ability:

- TD 337 for the transmission of: plant height, ear height and leaf area,
- TA 426 for the transmission of: plant height and ear height,
- TC 385 A for the transmission of: number of leaves above the ear and leaf area,
- TC 384 A for the transmission of: plant height and leaf area,
- TA 422 for the transmission of: leaf area,
- TC 399 for the transmission of: plant height, ear height and leaf area.

Differences between the inbred lines compared (Table 6) reflect the differences in the cumulative effects homozygous loci. The differences at the genetic level are demonstrated on the basis of correlation coefficients ( $r\hat{g}$ ).

As a result, the most pronounced difference in the level of gene expression of the additive takes place between different groups lines (A or B), (as is usual): TC 384 A - TA 428 ( $r\hat{g} = -0.93^{***}$ ), TC 398 - TA 426 ( $r\hat{g} = -0.91^{***}$ ), TC 398 - TD 337 ( $r\hat{g} = -0.86$ ), TC 384 A - TA 422 ( $r\hat{g} = -0.86$ ), TC 385 A - TA 422 ( $r\hat{g} = -0.72^{***}$ ).

The most pronounced degree of similarity in the additive gene expression between is between the inbred lines: TA 426 - TD 337 ( $r\hat{g} = 0.77$ ), TC 399 - TD 337 ( $r\hat{g} = 0.93$ ).

Table 6

Correlations at the level of additive genes corresponding to the inbred lines- parental genotypes - Turda 2013-2014

	TD337	TA426	TA428	TA422	TC385A	TC384A	TC398	TC399
TD 337	1							
TA 426	0.77	1						
TA 428	0.16	0.68	1					
TA 422	-0.55	0.10	0.64	1				
TC385A	0.04	-0.49	-0.48	-0.72	1			
TC384A	0.11	-0.53	-0.93	-0.86	0.68	1		
TC 398	-0.86	-0.91	-0.57	0.14	0.19	0.33	1	
TC 399	0.93	0.55	-0.12	-0.71	0.18	0.36	-0.74	1
P (5%)	0.71							
P (1%)	0.83							

### CONCLUSIONS

Research conducted to assess the relatedness/ differentiation of inbred lines may have important implications for increasing the maize breeding works efficiency, in the strategy of creating hybridization formulas.

Between the eight inbred lines studied existed phenotypic and genetic differences even between those originalted from the same initial material.

Variance analysis for cross diallel system showed distinct significant contribution for both years and hybrids in achieving plant height, ear height, the total number of leaves and leaf area.

The decomposition of orthogonal variance of the hybrids shows that for both crosses between lines from Comp. A and those from Comp.B there were significant differences for phenotypic traits (plant height, ear height, total number of leaves and leaf area).

Through comparative study between (A x A) and (B x B) hybrids there can be observed significant differences for plant height and ear height (A x A). For plant height and other phenotypic traits analyzed the difference between hybrids was in favor of those between AxB group.

Through the combining ability analysis of eight inbred lines, some distinguished by the ability to transfer several characters. Some valuable inbred are: TD 337, TA 426, TC 385 A, TC 384 A, TA 422, TC 399.

The most pronounced degree of differentiation at the level of additive gene expression result between inbred lines from different groups (A or B): TC 384 A - TA 428 ( $r_g = -0.93^{***}$ ), TC 398 - TA 426 ( $r_g = -0.91^{***}$ ), TC 398 - TD 337 ( $r_g = -0.86$ ), TC 384 A - TA 422 ( $r_g = -0.86$ ), TC 385 A - TA 422 ( $r_g = -0.72^{***}$ ).

The most pronounced degree of similarity for the additive gene expression result between the lines: TA 426 - TD 337 ( $r_g = 0.77$ ), TC 399 - TD 337 ( $r_g = 0.93$ ).

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