# **Original Paper**

## Classification of argentine maize landraces in heterotic groups

Carla Delucchi<sup>1\*</sup>, Guillermo H Eyhérabide<sup>1</sup>, Roberto D Lorea<sup>1</sup>, Daniel A Presello<sup>1</sup>, María E Otegui<sup>3</sup>, César G López<sup>2</sup>

<sup>1</sup>Instituto Nacional de Tecnología Agropecuaria, EEA Pergamino, Ruta 32 Km 4.5, 2700 Pergamino, Argentina <sup>2</sup>Universidad Nacional de Lomas de Zamora, Facultad de Ciencias Agrarias, Ruta 4 Km 2, 1836 Lavallol, Argentina <sup>3</sup>Departamento de Producción Vegetal, Facultad de Agronomía, Universidad de Buenos Aires, Av. San Martín 4453, C1417DSE Ciudad de Buenos Aires, Argentina.

\*Corresponding author: E-mail: cdelucchi@pergamino.inta.gov.ar

## Abstract

The genetic diversity of maize (*Zea mays* L) is a valuable and strategic natural resource that plays a key role in the breeding progress. However, exploitation of genetic variability from landraces has not reached a significant level of utilization in breeding programs in Argentina yet. In order to establish their breeding potential, the best 15 out of a group of about 300 landraces from Argentina, were evaluated for various agronomic characters in testcrosses with five lines representing different heterotic groups. Testcrosses were evaluated in nine environments during two growing seasons. A factorial array of those landraces and tester lines was used. Differences for landraces, testers, and landrace x tester interactions were detected for ear diameter and length, ear attachment and plant height, and grain yield. Yield data were further analyzed following additive main effects (landrace and tester) and multiplicative interaction (landrace x tester) models. The first two principal components were significant and accounted for 67% of that interaction. The first axis was consistent with the Argentine flint vs. US dent (Mo17), and US dent (B73) vs. US dent (Mo17) heterotic patterns. The second axis exhibited a contrast between Argentine flint and US dent (B73 or B73 derived line) heterotic groups. The first two principal components of the landrace x tester interaction and mean performance of testcrosses were considered to identify eight landraces as parents of three composite populations.

Keywords: genetic resources, heterotic groups, testcross evaluation, maize landraces

## Introduction

Maize (*Zea mays* L) shows a high genetic diversity (Brieger et al, 1958; Paterniani and Goodman, 1977; Goodman, 1983; Goodman and Brown, 1988). Several South American countries (Argentina, Bolivia, Brazil, Chile, Paraguay and Uruguay) have important collections of maize germplasm, many of them collected from farmers' fields in the 1970s, before generalized adoption of hybrid cultivars. In the case of Argentina, the main collection belongs to the National Institute of Agricultural Technology (INTA), and consists of 2365 accessions from 20 provinces located between 26° 07' S and 33° S (Hourquescos et al, 2005).

Several authors have emphasized that hybrid cultivars of maize adapted to temperate climates have a narrow genetic base (Zuber and Darrah, 1980; Stuber and Goodman, 1983; Smith et al, 1992). Stuber (1986) estimated that breeders took advantage of only about 5% of the available races of maize. While landraces were used as genetic resource of valuable inbred lines in the past, their present exploitation has not reached the potential impact on local breeding programs (Eyhérabide and Gonzalez, 1997).

Determination of heterotic groups and heterotic patterns is of fundamental importance in the improvement of maize hybrids. Heterotic groups have a strong impact in crop improvement because they

predetermine to a large extent the type of germplasm used in a hybrid breeding program for a long period of time (Melchinger and Gumber, 1998). The heterotic groups should not be considered as closed populations, and must be continuously extended by original germplasm introduction to ensure medium and longterm genetic gains from selection.

In Argentina, the heterotic pattern "Argentinian Orange Flint" (dark orange endosperm) vs. "US Yellow Dent" is used to develop inbred lines and hybrids (Maunder, 1992; Eyhérabide et al, 2006). Crosses between lines of both heterotic groups resulted in the release of highly productive hybrids. Heterotic groups "Reid Yellow Dent / Stiff Stalk Synthetic (BSSS)" and "Lancaster Sure Crop" were initially incorporated into elite flint lines through backcrosses (Brun and Dudley 1989). Since the final product of maize breeding programs in Argentina is the production of elite hybrids, an appropriate introduction of novel genetic variability requires its characterization in terms of combining ability (Eyhérabide and Gonzalez, 1997). In turn, relative performance in testcrosses with a group of divergent testers can be used as an estimation of genetic distance. According to Hallauer et al (1988), heterotic patterns among populations could be established based on the evaluation of testcrosses. This approach has been used extensively in corn (Nestares et al, 1999; Castañón et al, 1998; Betrán et al, 2003). Eyhérabide and Gonzalez (1997) evaluated the performance of 79 landraces with diverse testers to define heterotic groups of landraces based on effects of specific combining ability (SCA).

As part of a more extensive evaluation, 300 landraces from Argentina, belonging to the Genebank of INTA in Pergamino, were evaluated by crossing them with two types of broad-base testers, one of local origin and flint endosperm, and the other of North American origin and dent endosperm. Based on the combining ability for grain yield the best 15 landraces were selected for further studies (Hourquescos, Ferrer, Eyhérabide, personal communication). The objectives of this study were i) to evaluate the agronomic performance of the 15 selected maize landraces in crosses with inbred lines representative of different heterotic groups, ii) to assign the landraces to already established heterotic groups based on combining ability effects, and iii) to propose strategies of use of the selected landraces with the aim of broadening the base of the crop genetic variability.

#### Materials and Methods

The group of 15 landraces from Argentina mentioned in the introduction (Table 1) were testcrossed with five inbred lines: three US dent lines (B73, Mo17 and a Monsanto proprietary line coded as LE3), and two dark orange flint lines of Argentine origin (LP612 and LP122-2). B73 and Mo17 were developed by lowa State University and the University of Missouri, respectively; LE3 derives from B73 and B37 lines (Gerdes et al, 1993), and is widely used in temperate regions around the world; LP612 and LP122-2 are inbred lines developed by INTA and represent a flint x flint heterotic pattern of its breeding program. The single crosses and its parent inbred lines were considered representative of the heterotic patterns Reid Yellow Dent vs. Lancaster Sure Crop (B73 x Mo17, LE3 x Mo17), Argentine Flint A vs. Argentine Flint B (LP122-2 x LP612) and Argentine Flint (A or B) vs. US Dent (Reid Yellow Dent or Lancaster Sure Crop) (B73 x LP612, LE3 x LP612, Mo17 x LP612, B73 x LP122-2, LE3 x LP122-2, Mo17 x LP122-2).

During the 2005/2006 and 2006/2007 growing seasons, the single crosses between the five inbred lines, the landrace x tester testcrosses, and four check hybrid cultivars (ACA 2000, ALBION CL, DK 747 MG, and NIDERA 895), a total of 87 genotypes, were evaluated in experiments using a randomized incomplete block design (9 x 12 alpha-lattice design) with two replications. All the trials were planted in Pergamino on two sowing dates (referred to as Pergamino I and Pergamino II hereinafter), Ferré and Lomas de Zamora (Buenos Aires Province), and in Santa Isabel (Santa Fe Province), during the 2005/2006 season, and in Pergamino I and Pergamino II, Lomas de Zamora and Santa Isabel, during the 2006/2007 season. Seed of the single hybrid Mo17 x B73 was only available for the experiments on the 2006/2007 season, and the single cross B73 x LE3 was excluded from the evaluation due to insufficient quantity of seed. Sowing was adapted to the conventional system and zero-tillage by using the practices currently adopted by the farmers as well as fertilization and weed control practices. Each plot consisted in two rows of 5 m long with 70 cm between rows, resulting in a density of 70,000 plants ha<sup>-1</sup> approximately after thinning.

The check hybrids were not considered in the diallel analysis, but they were included in order to have a reference for the performance of elite cultivars. These checks were chosen so that they were representative of the range of agronomic performance and grain quality of the cultivars currently available in the market. The agronomic traits were evaluated in different environments as indicated in Supplementary Table 1.

Grain yield (Mg ha<sup>-1</sup>) was calculated on a 14% grain moisture basis. Kernel number per square meter

Table 1 - Landraces evaluated in testcrosses with five testers in nine environments, racial type and their site (Argentine Province) of collection. Code column indicates the number used to refer to each landrace in Figure 1.

Code	Landrace	Racial type	Site of collection
1	ARZM01042	"Cristalino Colorado"	Buenos Aires
2	ARZM01045	"Cristalino Colorado"	Buenos Aires
3	ARZM01073	"Cristalino Amarillo"	Buenos Aires
4	ARZM02003	"Cristalino Colorado"	Santa Fe
5	ARZM02023	"Dentado Amarillo"	Santa Fe
6	ARZM03014	"Amargo"	Entre Ríos
7	ARZM04062	"Crist. Amarillo Anaranjado"	Corrientes
8	ARZM06020	"Cristalino Colorado"	Chaco
9	ARZM07134	"Crist. Amarillo Anaranjado"	Formosa
10	ARZM14103	"Cristalino Colorado"	Córdoba
11	ARZM16008	"Dentado Amarillo"	Mendoza
12	ARZM16064	"Dentado Amarillo"	Mendoza
13	ARZM17035	"Cristalino Colorado"	San Luis
14	ARZM18017	"Cristalino Colorado"	La Pampa
15	ARZM18037	"Cristalino Colorado"	La Pampa

## classification of maize landraces

was estimated from the quotient grain yield / individual kernel weight. Individual kernel weight was taken on subsamples of 1,000 kernels per plot. Test weight was determined by indirect method using the automatic equipment TR400 (Tripette and Renaud, 400), from a sample of 500 grams of entire grains of maize. The cycle up to 50% of anthesis and 50% of silking were calculated as the thermal sum (in centigrade degrees day, °Cd, base temperature 8°C) across days from sowing to pollen shed and silking by 50% of plants in each plot. Ear height (cm) was the average height of 10 competitive plants measured from ground level to the node of top ear attachment. The plant height (cm) was the average height of those 10 plants, measured from the level of the soil up to the top end of the tassel. Both heights were measured 15 days after 50% anthesis. Root (plants leaning more than 30° from the vertical) and stalk (plants broken at or below the ear node) lodging were counted 10 days before harvest and expressed as percentages of final stands for each plot.

Given the percentages of relative efficiency of the analyses following the alpha-lattice design with regard to that of randomized complete blocks design (near to 100%), the statistical analyses were realized considering the latter design. Analyses of variance (ANOVA) of every individual experiment and combined across experiments for the different variables were performed using the Genes software (Aplicativo computacional em genética e estadística - www. ufv.br/dbg/genes/genes.htm). The landrace x tester testcrosses were considered as fixed effects and environments were considered random effects for the combined analyses of variance. The performance variability among testcrosses for all the evaluated traits was partitioned according to a factorial array of crosses between 15 landraces and five inbred lines, in order to evaluate the general combining ability of

Table 2 - Mean grain yields (Mg ha<sup>-1</sup>) of landrace x tester crosses, reference single hybrids and check hybrids across 8 environments (Pergamino I and Pergamino II, both in 2005/2006 and 2006/2007; Ferré 2005/2006; Lomas de Zamora 2005/2006 and 2006/2007; Santa Isabel 2005/2006).

Landrace		ML <sup>2</sup>				
	LP612	LP122-2	Mo17	B73	LE3	
 ARZM01042	7.67	8.71	7.23	7.75	8.48	7.97
ARZM01045	7.10	8.55	7.28	6.72	8.12	7.55
ARZM01073	8.09	8.02	7.42	7.37	7.45	7.67
ARZM02003	7.56	7.98	7.81	7.50	7.82	7.73
ARZM02023	7.31	7.34	6.98	6.59	6.75	6.99
ARZM03014	8.39	8.68	6.65	7.85	7.53	7.82
ARZM04062	7.81	7.46	6.76	7.70	8.33	7.61
ARZM06020	8.12	7.49	8.64	7.91	7.99	8.03
ARZM07134	7.91	7.78	8.04	7.71	8.12	7.91
ARZM14103	7.44	8.56	8.40	8.78	8.09	8.25
ARZM16008	7.85	8.67	7.58	7.61	8.10	7.96
ARZM16064	7.55	8.49	6.64	8.55	7.85	7.81
ARZM17035	7.08	7.88	7.71	7.79	8.24	7.74
ARZM18017	7.45	8.00	7.78	8.14	8.22	7.92
ARZM18037	7.56	8.39	6.94	7.51	8.36	7.75
MT <sup>1</sup>	7.66	8.13	7.46	7.70	7.96	
Line Tester						Mean
LP612		8.10	6.45	8.10	8.13	7.70
LP122-2	8.10		8.76	9.22	8.77	8.71
Mo17	6.45	8.76		-	7.94	7.72
B73	8.10	9.22	-		-	8.66
LE3	8.13	8.77	7943	-		8.28
Mean	7.70	8.71	7.72	8.66	8.28	
Checks						
NIDERA895						8.62
ALBION CL						7.74
DK 747 MG						9.19
ACA 2000						8.35

<sup>1</sup>MT: means of testers across landraces.

<sup>2</sup>ML: means of landraces across testers.

LSD 0.05 between the means of two landrace x tester crosses: 0.86 Mg ha-1

LSD 0.05 between the means of two reference single crosses: 0.83 Mg ha<sup>-1</sup>

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every group of progenitors (landraces and testers) and the specific combining ability between them. Phenotypic correlation coefficients between grain yield and its components evaluated across five environments (Pergamino I and Pergamino II 2005/2006 and 2006/2007, and Ferré 2005/2006) were also estimated.

On the other hand, the interactions between landraces and testers were further analyzed following additive main effects and multiplicative interaction (AMMI) models (Gauch and Zobel 1988; Crossa et al, 1990). The mean squares for landraces and testers correspond to the variation for their respective general combining ability (GCA effects, and the landrace x tester mean square corresponds to the variation for specific combining ability (SCA) effects. In turn, the variation associated with the specific combining ability effects could be divided into principal components according to the AMMI models in order to use this information to classify landraces in heterotic groups (Eyhérabide and Gonzalez, 1997; Betrán et al, 2003). Analyses of AMMI models were performed using a program written in SAS (SAS Institute, 1999; Vargas and Crossa, 2000). Data matrix was a 15 x 5 grain yield array (15 rows for landraces x 5 columns for tester lines). In such a way, the landrace x tester interaction sum of squares was partitioned into principal components axes, and the singular value, the landrace eigenvector and the tester eigenvector for each axis determined. Gollob's test (Gollob, 1967) was used to determine the significance of each principal component axis. Multiplying the elements of the eigenvectors by the square root of the corresponding eigenvalues, the tester scores and the landrace scores for the n principal component were obtained. Their product provides an estimation of the expected specific interaction value for each landrace x tester combination.

Additionally, the landrace scores for the first three principal component axes obtained from the AMMI analysis were used as the variable in a cluster analysis. This was carried out by means of the procedure PROC CLUSTER (SAS Institute, 1999) based on the Ward's (Ward, 1963) minimum variance method. The more appropriate number of clusters was defined according to the statisticians Pseudo F (Calinski and Harabasz, 1974) and Pseudo t<sup>2</sup> (Duda and Hart, 1973).

#### **Results and Discussion**

Mean grain yield for the landrace x tester testcrosses across the eight environments varied from  $6.59 \text{ Mg ha}^{-1}$  (ARZM02023xB73) to  $8.78 \text{ Mg ha}^{-1}$ (ARZM14103 x B73) (Table 2). The best fourteen crosses did not show significant differences with the commercial check DK 747 MG, which was the check hybrid that showed the highest grain yield across environments. Considering Santa Isabel 2005/2006, the highest yield environment, grain yield of the Table 3 - Partial diallel analysis combined across eight environments (Pergamino I and Pergamino II, both in 2005/2006 and 2006/2007; Ferré 2005/2006; Lomas de Zamora 2005/2006 and 2006/2007; Santa Isabel 2005/2006) for grain yield (kg ha<sup>-1</sup>).

Source of variation	DF	Mean Square
Blocks/Environments	8	5682706
Genotypes <sup>1</sup>	86	5719237**
Testcrosses	74	4756159**
Landraces GCA	14	6324973**
Testers GCA	4	17003244*
Landrace x tester SCA	56	3489163*
Reference and check hybrids	12	11658218**
Environments	7	342108614**
Genotypes <sup>1</sup> x Environment	602	3578602**
Testcrosses x Env.	518	2803384**
Landraces GCA x Env.	98	2359217**
Testers GCA x Env.	28	6404735**
Landrace x tester SCA x Env.	392	2657186**
Reference and check hybrids x Env.	84	8359113**
Pooled Error	688	1568470
Total	1391	
General Mean (kg ha-1)		7851
CV (%)		16,0

<sup>1</sup>Landrace x tester testcrosses, reference single hybrids and check hybrids. \*\*, \*: Significant at the 0.01 and 0.05 probability levels, respectively.

landrace x tester crosses varied from 7.54 Mg ha<sup>-1</sup> (ARZM01045 x B73) to 13.64 Mg ha<sup>-1</sup> (ARZM18037 x LE3) (data not shown). At this environment, 73 of the 75 testcrosses evaluated did not show significant differences with regard to the best commercial check, ACA 2000.

The combined analysis of variance across environments for grain yield (Table 3) detected significant differences (P<0.01) between the genotypes (testcrosses, reference single hybrids and check hybrids), with a general mean of 7.85 Mg ha<sup>-1</sup>. The genotype x environment interaction was also highly significant (P<0.01). Partitioning of the "Genotypes" sum of squares showed also highly significant differences between the 75 testcrosses (P<0.01, Table 3). In addition, partitioning of the testcrosses source of variation according to a factorial array revealed significant differences for grain yield (P<0.01) between landraces (GCA effects), between testers (GCA effects; P<0.05) and, at a lower significance level (P<0.07), between landrace x tester (SCA effects). These results indicate the presence of genes with both, additive and nonadditive effects responsible for the genetic variability among landrace x tester crosses. The testcross x environment interaction and both, the general and specific combining abilities x environment interactions were highly significant (P<0.01).

Phenotypic correlation between grain yield and its components (Supplementary Table 2) evaluated across five environments (Pergamino I and Pergamino II 2005/2006 and 2006/2007, and Ferré 2005/2006) was only significant (P<0.01) between kernel number per square meter and grain yield (r=0.64). On the other hand, kernel weight and kernel number showed



Figure 1 - Biplot of the AMMI analysis of 15 selected landraces (Identification numbers in Table 1) and five testers for grain yield across environments.

a negative association (r=-0.36, P<0.01). There are reported evidences about the positive association between grain yield of commercial hybrids in Argentina and their kernel weight (Cirilo and Andrade, 1994; Otegui et al, 1995). However, in coincidence with the findings of the present research, literature is consistent indicating kernel number as the principal determinant of grain yield in maize (Bolaños and Edmeades, 1996; Edmeades et al, 1999; Andrade et al, 1999). The negative association between kernel number and kernel weight coincides with previous results evaluating commercial hybrids of Argentina (Borrás and Otegui, 2001) and it has already been widely discussed in relation with its ecophysiological bases (Gambín et al, 2006).

Highly significant differences (P<0.01, Supplementary Table 3) among testcrosses were detected for all the variables evaluated in the combined analysis of variance across environments except for test weight (not presented). According to the combined diallel analysis, landraces GCA effects were significant (P<0.01 and P<0.05) for all traits evaluated, except for kernel weight. Tester lines GCA effects were also significant (P<0.01 and P<0.05) for all traits except for anthesis-silking interval and stalk lodging. SCA effects for landrace x tester crosses were highly significant (P<0.01) for ear length and diameter, number of kernel rows, ear and plant height. Kernel weight and number per square meter, cycle up to 50% of anthesis and silking, anthesis-silking interval and stalk lodging did not show significant differences in effects of landrace x tester SCA (Supplementary Table 3).

AMMI models for grain yield across environments were applied in order to classify landraces in heterotic groups (Eyhérabide and Gonzalez, 1997; Betrán et al, 2003). More than a half of the total sum of squares (56%) accounted for the full model for grain yield across environments corresponded to the interaction landrace x tester sum of squares. This sum of squares was divided in principal components, whose first two axes accounted for about 67% of it (43.8% and 23.2% for the first and second axis, respectively). In the corresponding biplot (Figure 1), testers resulted located in different quadrants according to its genetic origin. For the first axis, the two largest scores (absolute values) for the testers corresponded to the flint line LP122-2 (-0.85) and to the dent line Mo17 (1.24). Moreover, according to the respective vector angle (about 180°) there is a correlation between their testcrosses. The other dent line testers, B73 and LE3, as well as the flint tester LP612, had negative and closed to zero scores (-0.08, -0.14 and -0.18, respectively). In addition, the vector angle between B73 and LE3 is consistent with their genetic relationship. Thus, the first axis could be interpreted as indicative of heterotic patterns, or contrast, between germplasm groups composed by i) Argentine Flint A or B (LP122-2 or LP612) x US dent (Mo17), and ii) Reid Yellow Dent (BSSS-B73 or LE3) x Lancaster Sure Crop (Mo17). For the second axis, local flint testers had positive or closed to zero scores (LP612: 1.04; LP122-2: -0.01), whereas US dent testers had negative or closed to zero scores (B73: -0.59; LE3: -0.49 and Mo17: 0.05). These results were consistent with the existence of heterotic patterns made up by Argentine flint B (LP612) x US dent (BSSS-B73 or LE3), and in a lesser extent Argentine Flint A (LP122-2) x US dent (BSSS-B73 or LE3). B73 and LE3 scores across the two axes resulted consistent with the genetic relationship of these SSS lines.

According to Eyhérabide and Gonzalez (1997) evaluated landraces could be classified on the basis of the sign and magnitude of their respective interaction with testers of different genetic background. As an example, contribution of landrace x tester interaction corresponding to landraces with positive scores for the second principal component would be favorable to grain yield of testcrosses with flint tester LP612 and in a lesser extent with tester Mo17, because these landraces and tester scores, have the same sign, but it would be detrimental to grain yield of testcrosses with dent testers LE3 and B73,





Figure 2 - Dendrogram for Ward's minimum variance cluster analysis of 15 landraces according to their scores for the first three principal components of the landrace x tester interaction of the AMMI analysis for grain yield across environments.

Table 4 - Means of the selected landraces across environments and testers for cycle up to 50% of anthesis (GDDA, growing degree days to anthesis), kernel number per square meter (KN m<sup>-2</sup>), ear height (EH), percentage of stalk lodging (SL), mean G.C.A. effects (GCA) and grain yield.

								Grain yield for testers (Mg ha-1)4						
	Landrace	GDD1 (°Cd)	KN m <sup>-2 2</sup>	EH (cm) <sup>2</sup>	%SL <sup>3</sup>	GCA <sup>4</sup>	LP122-2	LP612	B73	LÉ3	Mo17	Mean		
Composite population 1														
	ARZM03014	996.1	2372.2	120.9	19.7	37.5	8.68	8.39	7.85	7.53	6.65	7.82		
	ARZM16008	984.3	2441.2	107.8	16.5	179.1	8.67	7.85	7.61	8.10	7.58	7.96		
	Mean	990.2	2406.7	114.4	18.1	108.3	8.67	8.12	7.73	7.81	7.11	7.89		
Composite population 2														
	ARZM16064	1003.3	2376.2	127.5	25.1	32.3	8.49	7.55	8.55	7.85	6.64	7.81		
	ARZM01042	975.2	2505.5	118.3	14.0	188.1	8.71	7.67	7.75	8.48	7.23	7.97		
	ARZM14103	954.9	2480.9	120.9	19.0	470.6	8.56	7.44	8.78	8.09	8.40	8.25		
	ARZM18017	967.5	2379.8	114.0	12.8	135.2	7.99	7.45	8.14	8.22	7.78	7.92		
	Mean	975.2	2435.6	120.2	17.7	206.55	8.44	7.53	8.30	8.16	7.51	7.99		
Composite population 3														
	ARZM06020	975.2	2318.6	120.3	16.1	246.6	7.49	8.12	7.91	7.99	8.64	8.03		
	ARZM07134	970.5	2482.9	116.5	17.3	129.7	7.78	7.91	7.71	8.12	8.04	7.91		
	Mean	972.8	2400.8	118.4	16.7	188.15	7.64	8.01	7.81	8.05	8.34	7.97		

<sup>1</sup>Data from Pergamino I and II 2005/2006, and Pergamino I 2006/2007.

<sup>2</sup>Data from Pergamino I and II 2005/2006, Ferré 2005/2006, and Pergamino I and II 2006/2007.

<sup>3</sup>Data from Santa Isabel 2005/2006, and Pergamino I and II 2006/2007.

<sup>4</sup>Data from Pergamino I and II 2005/2006, Ferré 2005/2006, Lomas de Zamora 2005/2006, Santa Isabel 2005/2006, Pergamino I and II 2006/07, and Lomas de Zamora 2006/2007.

because landrace and dent tester scores have opposite sign. Contribution of landrace x LP122-2 interaction to grain yield would be negligible since LP122-2 score for the second axis is near zero.

Crosses between landraces and testers whose scores for the first two principal components for grain yield are located in the same guadrant (Figure 1) would benefit from a favorable contribution to their grain yield in terms of the first and second principal components of the interaction. On the other hand, the contribution would be unfavorable for crosses between landraces and testers whose scores are in different quadrants on the same diagonal. The final contribution of the interaction of other landrace x tester combinations to grain yield will depend on the algebraic sum of the products between the respective scores in both axes (Eyhérabide and Gonzalez, 1997). Contribution of the landrace x line interaction to the performance of testcrosses would be of minor importance for those landraces whose scores are close to zero for both axes; such was the case of landraces ARZM04062 (7) and ARZM01045 (2). On the contrary, landraces ARZM06020 (8) and ARZM03014 (6) could have, through its effect on the first principal component a larger contribution to the testcross performance when crossed to Mo17 and LP122-2, respectively.

Ward's cluster analysis of the 15 landraces, on the basis of their scores for the first three principal components of the landrace x tester interaction (the third axis explained 20,5% of the variation, in total approximately about 87% for the first three principal components) allowed the identification of four groups (Figure 2). Considering the site of collection of landraces (Table 1) and assignment to clusters suggests that the geographical site of collection was not related to their clustering.

Mean grain yield of landraces in testcrosses above the average (Table 2), positive values of GCA, and interaction with testers obtained from the AMMI models (Figure 1), provided a criteria for the preliminary selection of eight landraces to be used as parents of three diverse populations (Table 4). ARZM03014 (6) and ARZM16008 (11) showed favorable interactions with flint testers LP122-2 and LP612, thus, they could be intermated to form one population. Landraces ARZM16064 (12) and ARZM01042 (1), ARZM14103 (10) and ARZM18017 (14) exhibited favorable interactions with dent testers LE3 and B73, both corresponding to the Reid Yellow Dent-BSSS heterotic group, and could be intermated to form a second composite population. Finally, ARZM06020 (8) and ARZM07134 (9) combined favorably with the dent tester Mo17, representative of the heterotic group Lancaster Sure Crop, hence, they could be intermated to form a third heterogeneous population. If the members included in each of the four clusters obtained through the Ward's method (Figure 2) are examined, it is possible to distinguish that the landraces assigned to the first two composite populations, resulted included in different clusters than those assigned to the third population. These ones were grouped in the same cluster, showing partial correspondence between the results obtained from the biplot of the AMMI analysis and from the clustering analysis according to their scores for the first three principal components of the landrace x tester interaction.

The correspondence evidenced between the different sources of information show the utility of AMMI models in the analysis of landrace x tester testcrosses, since generally they are used in the evaluation of genotype x environment interactions. The information provided by the principal components obtained according to the AMMI models was extremely useful in the classification of the landraces evaluated in heterotic groups, because it allowed relating them directly to the tester lines. On the other hand, the grouping originated by the hierarchical cluster analysis arises from using a singular value decomposition of SCA effects between the landraces and the five tester lines. The results of this study suggest, therefore, the convenience of using AMMI models when it is necessary to evaluate germplasm in relation to different heterotic groups and patterns in use.

In previous studies, using estimators of the relative number of favorable alleles in the landraces, which are absent in a reference hybrid (Dudley, 1987; 1988), Lorea et al (2008) evaluated the same landraces analyzed in this work for grain yield. Reference hybrids were LP612 x LP122-2, LP612 x Mo17, and LP122-2 x Mo17. Most of the landraces presented favorable alleles to improve the performance of the reference hybrids. In relation to the first hybrid the most promising varieties would be ARZM01042 and ARZM06020; ARZM16008 and ARZM16064 also showed a high frequency of favorable alleles. For the other two hybrids ARZM07134 showed the highest concentration of favorable alleles and similarity with LP612 and LP122-2, so it could represent a new heterotic group to exploit with good combination with dent lines, related to Mo17.

According to the results, more than one strategy could be considered to use the proposed composite populations as a germplasm source for the development of inbred lines. In all instances, carrying out a scheme of recurrent selection to improve grain yield and to reduce root and stalk lodging would be desirable. A scheme of half-sib intrapopulation recurrent selection could be followed using the tester that contrasts with each defined heterotic group. Thus, individuals from the composite population derived by intermating landraces ARZM03014 and ARZM16008, could be self pollinated and selected based on the performance of their testcrosses with LP612 or LP122-2. An alternative to this strategy consists of intermating landraces of one composite population with testers that demonstrated minor genetic distance (the ones that presented scores of different sign in the AMMI biplot). The introduction of elite variability would improve the performance of composites for standability, and would increase probabilities of obtaining new promising inbred lines in a smaller number of cycles of selection. Then, during the stage of development of inbred lines from this improved composite population, evaluation for combining ability should be performed using lines from opposite heterotic groups (those that had similar signs to landraces in the AMMI biplot).

As it was mentioned before, there was a partial

correspondence between the assignment of landraces to the three composites and results from the cluster analysis. If just two clusters were considered, two composite populations could be proposed for practical reasons. Landraces to be included in Composite A would be those with positive scores for the first principal component [ARZM07134 (9), ARZM06020 (8), ARZM14103 (10), ARZM18017 (14)], and those to be included in Composite B would be those with negative scores for the first principal component [ARZM01042 (1), ARZM16008 (11), ARZM03014 (6), ARZM 16064 (12)]. Mo17 could be introduced in Composite B and LP122-2 in Composite A. Reciprocal recurrent selection could be utilize to improve both composites, and to derive new inbred lines that should be tested by combining ability preliminarily using Mo17 (inbred families from Composite A) and LP122-2 (inbred families from Composite B). This alternative would be more efficient in terms of resources and coupling with a program of hybrid development. Independently of the chosen strategy, the evaluated landraces showed potential to broadening the base of the crop genetic variability.

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