

Six cycles of S₁ recurrent selection in two Spanish maize synthetics

J. I. Ruiz de Galarreta^{1*} and A. Álvarez²

¹ NEIKER. Centro Arkaute. Apdo. 46. 01080 Vitoria. Spain

² Estación Experimental de Aula Dei. CSIC. Apdo. 202. 50080 Zaragoza. Spain

Abstract

European flint × U.S. dent is the most common heterotic pattern used in early maize (*Zea mays* L.) breeding programs in Spain. Two synthetic maize populations, EZS1 (formed from four Spanish flint populations), and EZS2 (formed from four American dent populations), were developed in Zaragoza, Spain. These synthetics, which showed heterotic behaviour, were subjected to six cycles of S₁ recurrent selection for increased grain yield and reduced lodging. The aim of this study was to investigate the effectiveness of selection on these and other agronomic traits in both synthetics. The original populations, EZS1C0 and EZS2C0, and the six cycles of selection of both, were evaluated at two locations over two years. Over the six selection cycles, the average yield of EZS1 and EZS2 increased by 0.82 Mg ha⁻¹ and 0.93 Mg ha⁻¹ per cycle respectively, while lodging decreased by 2.76% and 2.44% per cycle respectively. Changes due to selection were significant for all test traits in both synthetics. It was concluded that S₁ selection is effective in improving the yield of, and reducing lodging in, both synthetics.

Additional key words: heterotic pattern, local populations, recurrent selection, *Zea mays* L.

Resumen

Seis ciclos de selección recurrente S₁ en dos sintéticos de maíz españoles

El patrón heterótico de maíz (*Zea mays* L.) más utilizado en España, en programas de mejora genética, es el formado por el germoplasma liso europeo y el dentado de origen norteamericano. En Zaragoza, España, se obtuvieron dos poblaciones sintéticas de maíz, EZS1 (formada por cuatro poblaciones lisas españolas), y EZS2 (formada por cuatro poblaciones dentadas americanas). Estas poblaciones mostraron un buen comportamiento heterótico inicial y fueron sometidas a seis ciclos de selección recurrente intrapoblacional, por el método de familias S₁, para mejora de rendimiento y de encamado de planta. El objetivo de este trabajo fue estudiar la eficacia de la selección sobre el rendimiento y otros caracteres agronómicos en ambas poblaciones sintéticas. Las poblaciones originales (EZS1C0 y EZS2C0) y los seis ciclos de selección de ambas fueron evaluadas en dos localidades durante dos años. En el conjunto de los seis ciclos, y para el carácter rendimiento, el incremento promedio por ciclo fue de 0,93 Mg ha⁻¹ en EZS1 y de 0,82 en EZS2. El encamado de planta decreció 2,76% y 2,44% , por ciclo, respectivamente. Las ganancias genéticas debidas a la selección fueron significativas en ambas poblaciones y para todos los caracteres. Se puede concluir que en ambas poblaciones la selección por el método de familias S₁ fue eficaz para la mejora del rendimiento y la disminución del encamado.

Palabras clave adicionales: mejora genética, patrones heteróticos, poblaciones locales, *Zea mays* L.

Introduction

Maize hybrid production involves the use of inbreds derived from distantly related groups of breeding materials, but which combine well and produce better-performing progeny (Mumm and Dudley, 1994). Most maize hybrid seed in the USA is derived from the Corn

Belt dent race, but only a small portion of its available variation is used (Goodman and Brown, 1988). Heterosis and heterotic pairings are of central interest in maize breeding (Hallauer, 1990). Ordás (1991) studied the heterotic relationship between Corn Belt dent populations and Spanish flint germplasm. Most of the maize cultivars in Spain are hybrids based on this heterotic pattern (Moreno-González *et al.*, 1997), the limited genetic diversity of which has been demonstrated by Smith (1989), at least for the hybrids grown

* Corresponding author: jiruiz@neiker.net
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in France. It is therefore desirable to expand the genetic base of European cultivated maize by introducing new germplasm.

Recurrent selection based on S_1 progenies (Eberhart, 1970) is a good method of achieving improvement within populations (Moll and Smith, 1981) and has been proposed as a particularly promising means of improving yields. Moreno-González and Cubero (1993) reported increased grain yields in 19 selection experiments based on the evaluation of selfed S_1 or S_2 maize families. These increases averaged 3.56% per cycle in the populations *per se*, and 1.80% in the population testcrosses. However, several studies based on experimental data suggest that genetic variability in such populations becomes reduced after the initial cycles (Tanner and Smith, 1987), limiting opportunities for selection.

The maize breeding program at the Aula Dei Experimental Station, Zaragoza (Spain), developed synthetic populations based on the knowledge of the heterotic behaviour of yield components (e.g. number of ears, etc.). The development of two synthetics, EZS1 (made by intercrossing Spanish populations) and EZS2 (formed from North American populations), began in 1982. Preliminary results indicated that superior hybrids might be developed from the cross between these synthetics (Álvarez *et al.*, 1993). As the initial yields of both synthetics were low, they were separately subjected to an S_1 progeny recurrent selection scheme for six cycles, in order to render them competitive genetic contributors to the maize breeding program. Cycles 0, 1 and 2 (C0, C1, C2) of EZS1 and EZS2 have been previously evaluated for their response to several selection traits (Garay *et al.*, 1996a), as well as to determine their ability to combine (Garay *et al.*, 1996b). Least squares regression analysis based on the Smith (1983) model confirmed that both populations showed increased grain yield (Garay *et al.*, 1996a).

The aims of the present study were to evaluate the changes in the synthetic populations EZS1 and EZS2 after six cycles of S_1 intrapopulation selection, and to analyse future breeding strategies for the continued improvement of both populations.

Material and Methods

Plant materials

The EZS1 synthetic was formed by crossing four old Spanish, open-pollinated landraces with diverse

geographic origins and agronomic characteristics: 'Amarillo de Utrera', 'Hembrilla' (both described by Sánchez-Monge, 1962), 'Fino', and 'Hembrilla/Queixalet'. The EZS2 population was derived from crosses between the populations AS3-(HT)C3 (Peterson *et al.*, 1976), BS3 (Hallauer *et al.*, 1974), BS17, and BS1(HS)C1 (Russel, 1979), which combine a wide diversity of North American germplasm improved for one or more traits (mainly grain yield, and resistance to corn borer, *Ostrinia nubilalis* Hübner). Crosses to form the new synthetics were made in 1982 and 1983. The synthetics were later intermated twice, and labelled as either the C0 or original populations.

Between 1986 and 1991, both C0 populations were subjected to two cycles of recurrent selection based on S_1 progeny performance. In each cycle, a rank summation index for each progeny was calculated as the sum of the ranks for grain yield (adjusted to 140 g kg⁻¹ moisture) and percentage lodging. The 10 progenies with the lowest index values and with grain moisture below the mean were selected. The original 100 progenies were pre-selected for plant health, ear size and shape, and seed weight from about 250 selfed plants per population. Remnant S_1 seeds of the 10 selected families were used for recombination. Seven populations were eventually obtained for EZS1 and EZS2 (C0-C6). The seeds for the evaluation trials were obtained in the same year under the same conditions. A minimum of 150 plants per cycle were chosen for multiplication. Three hybrid maize controls were included in the trials in order to verify the correct functioning of the vegetative cycle, but these were not considered in the final analyses.

Evaluation trials

Post-selection progress was tested over two years (2002 and 2003) at two locations: Montañana (41°44'30"N, 0°47'00"W, 218 m altitude) and Peñaflor (41°46'32"N, 0°50'00"W, 225 m altitude) in the Province of Zaragoza, Spain. The trials were flood irrigated throughout the growing seasons to supplement the low natural rainfall (240 and 180 mm during the 2002 and 2003 growing seasons respectively). The 2002 trial plots received eight irrigations of about 50 mm each, while the 2003 plots received 10 irrigations. Cultivation operations, fertilization, and pest and weed control were undertaken according to local practice. All trials plants were machine-planted in mid-May in both years, with a stand density of 66,000 plants ha⁻¹. Each plot consisted

of two 5 m rows spaced 0.75 m apart. The entire plot was harvested manually in mid-November in both years. The experimental design for all trials involved a randomised complete block with three replicates. The data recorded for each plot were: days to silking, grain moisture at harvest (g H₂O kg⁻¹), lodging (estimated as the percentage of plants showing either root or stalk lodging), and yield (weight of grain expressed as Mg ha⁻¹, adjusted for a kernel moisture of 140 g H₂O kg⁻¹).

ANOVA was performed for each of the four environments (2 localities, 2 years) according to the randomised complete-block design. A combined analysis of variance across environments was then performed; the environments were considered to be random and the populations fixed. Means were compared using the LSD test (Steel and Torrie, 1980). All calculations were performed using the SAS package (SAS, 2000). Simple correlation coefficients among averaged traits over replications and years were calculated for each population. The linear regression coefficients obtained from the regression models were used as estimates of the average rates of response per cycle. Linear regression and correlation coefficients were obtained using the PROC GLM and PROC CORR procedures, respectively, of the same statistical software (SAS, 2000).

Results

The combined analyses of variance over years and locations (data not shown) revealed highly significant ($P \leq 0.01$) differences among entries for all traits except for grain moisture (significant at $P \leq 0.05$). The interaction mean square was used as the error term since the *genotype x environment* interactions were significant for all traits. Grain yield and lodging resistance were the two selection criteria used in population improvement. The selection response for these traits can be considered direct, whereas the responses of other traits must be regarded as correlated.

Means were compared among cycles across the four environments (Table 1). Highly significant ($P \leq 0.01$) linear increases in grain yield and reductions in lodging were seen in both populations over the selection cycles, as well as significant differences ($P \leq 0.05$) in grain moisture. The two synthetics obtained after the six cycles of S₁ selection showed significantly better grain yields and less lodging than the original populations EZS1C0 and EZS2C0 (Table 2). A good response to indirect selection was also observed for grain moisture in both synthetics. In EZS2, grain yield increased linearly during the S₁ recurrent selection program improving

Table 1. Means of grain yield and other agronomic traits in the two synthetic maize populations (EZS1 and EZS2) over six cycles of S₁ progeny recurrent selection

Population and selection cycle	Grain yield (Mg ha ⁻¹)	Silking (Days)	Lodging (%)	Grain moisture (g H ₂ O kg ⁻¹)
EZS1C0	3.57f	79.2b	18.8a	230a
EZS1C1	5.41e	79.7b	17.0a	232a
EZS1C2	6.62d	80.2ab	9.9b	214b
EZS1C3	6.95cd	80.9ab	8.8b	204bc
EZS1C4	7.33c	81.0ab	7.6b	198c
EZS1C5	8.00b	81.1ab	3.8c	199c
EZS1C6	8.52a	82.6a	2.9c	202bc
LSD (0.05) ¹	0.49	1.9	2.6	13
EZS2C0	3.26e	81.0b	15.2a	236a
EZS2C1	5.98d	81.1b	14.8a	248a
EZS2C2	7.08c	83.6a	8.8b	221b
EZS2C3	7.55c	83.1a	7.4bc	206cd
EZS2C4	7.56c	82.9ab	5.7cd	208bcd
EZS2C5	8.58b	83.1a	5.6cd	197d
EZS2C6	9.34a	82.9ab	3.5d	214bc
LSD (0.05)	0.42	1.9	2.4	14

¹ LSD: least significant difference between cycles of selection ($P \leq 0.05$). Means followed by the same letter do not differ significantly ($P \leq 0.05$).

Table 2. Estimates of regression coefficients (b), intercepts (I) and coefficients of determination (R^2) in the selection response of four traits to six S_1 recurrent selection cycles in the two maize synthetics (EZS1 and EZS2)

Trait	EZS1			EZS2		
	b	I	R^2	b	I	R^2
Grain yield ($Mg\ ha^{-1}$)	0.82**	4.37	0.90	0.93**	4.45	0.93
Silking (days)	0.42**	79.3	0.80	0.52**	81.1	0.74
Lodging (%)	-2.76**	18.3	0.90	-2.44**	14.8	0.90
Grain moisture (%)	-0.58**	222	0.60	-0.70**	229	0.66

** $p \leq 0.01$.

0.93 $Mg\ ha^{-1}$ per cycle. In EZS1 the increase in yield per cycle was 0.82 $Mg\ ha^{-1}$. Lodging was reduced to similar extents in both the EZS1 and EZS2 populations (-2.76 and -2.44 % per cycles respectively). Grain moisture was also reduced in both populations, with similar values recorded (-0.43 and -0.47 % for EZS1 and EZS2 respectively). The number of days to silking increased (an undesirable effect) in both synthetics (0.52 d in EZS2 and 0.42 d. in EZS1 per cycle).

Figures 1A and 1B show the yield and lodging responses for the original populations from C0-C6 respectively. Grain yield increased linearly with the selection procedure. EZS2 showed a higher grain yield than EZS1 in all selection cycles.

The correlation coefficients between grain yield and both silking and lodging were significant ($r = 0.92$ and $r = -0.98$ respectively; $P \leq 0.01$) for EZS1C0 (Table 3). In the same population, grain moisture and yield were also significantly correlated ($r = -0.86$; $P \leq 0.05$). In the synthetic EZS2C0, neither of the correlations between yield and either silking or grain moisture were significant.

However, the correlation coefficient between yield and lodging was significant ($r = -0.92$; $P \leq 0.01$).

Discussion

S_1 recurrent selection was effective in improving both EZS1 and EZS2 over six cycles of selection. Some authors have reported the effectiveness of recurrent selection-based S_1 families, e.g., Rodríguez and Hallauer (1988), who investigated ten populations, and Ceballos *et al.* (1991) who compared cycles of full-sib and S_1 family selection in subtropical populations of maize. Butrón *et al.* (2002) showed the effectiveness of intrapopulation recurrent selection in improving resistance to corn earworm (*Helicoverpa zea* Boddie).

Ten S_1 progenies were used to form the selected populations in each selection cycle, the usual practice for avoiding notable genetic drift (Valés *et al.*, 2001). Weyhrich *et al.* (1988) indicate that drift is a stronger force than selection in terms of changes to allele fre-

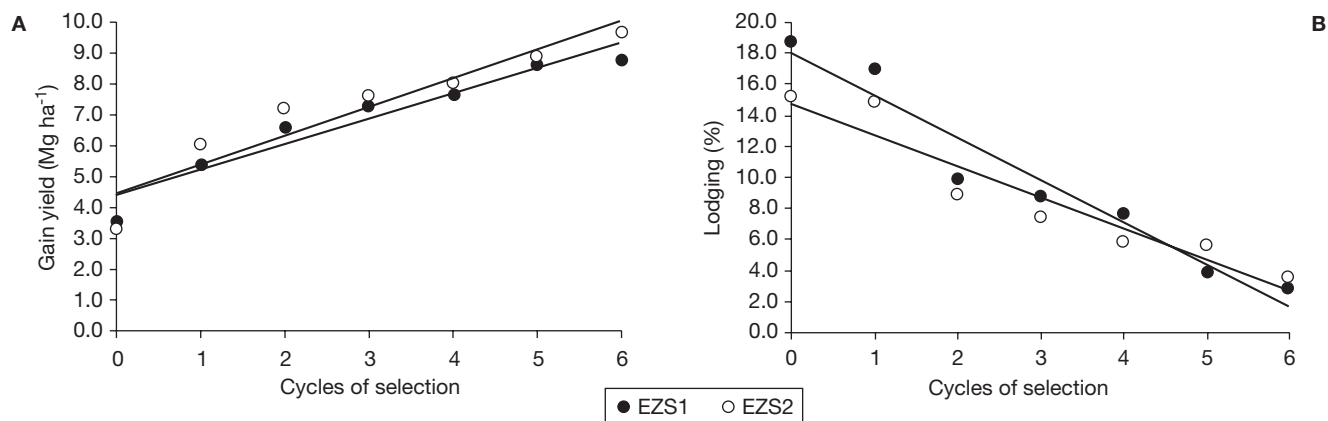


Figure 1. A) Grain yield, and B) lodging of the original cycle (C0) and all populations obtained after six cycles of S_1 recurrent selection in the two maize synthetics.

Table 3. Simple correlation coefficients between grain yield and other agronomic traits averaged over replications and years in the two maize synthetics over six cycles of recurrent selection

Population	Silking	Lodging	Grain moisture
EZS1C0	0.92**	-0.98**	-0.86*
EZS2C0	0.55	-0.92**	-0.73

*, ** $P \leq 0.05$ and $P \leq 0.01$, respectively.

quencies when fewer than 10 lines are recombined. Changes in agronomic traits other than the grain yield selection target were variable in similar breeding programs that used S_1 recurrent selection schemes (Garay *et al.*, 1996a). In the present study, most of the changes were in the desired direction, except for days to silking, which significantly increased by 3 and 2 days in the sixth selection cycle compared to the original populations of EZS1 and EZS2 respectively. Therefore, increases in grain yield were associated with a longer time to flowering, although the vegetative cycle of both populations still remained in the range acceptable for the conditions of the Zaragoza area. Similar results were reported by Garay *et al.* (1996a) who performed two selection cycles in the same populations.

Grain yield increases per cycle were larger than those obtained in other studies (Oyervides-García and Hallauer, 1986; Ordás, 1991; Valés *et al.*, 2001). High genetic gains was seen for both populations, especially in the first cycle. These may have been due to the heterogeneity of the synthetic material, as well the high selection index (10%) used with the original populations in the first selection cycles. In addition, some of the open-pollinated landraces forming the synthetics had different geographic origins and agronomic characteristics, increasing their heterogeneity; these landraces have never been genetically improved.

The future management of these populations should attempt to increase the selection pressure against high grain moisture in both synthetics. S_1 progeny recurrent selection for grain yield should continue in both populations since a very favourable linear response is obtained. However, it would not be advisable to do this without paying attention to maturity. An interpopulation recurrent selection method may improve the result of population crosses, and at the same time find the best cross combinations between selected families for consolidating the heterotic pattern between Corn Belt dent and Spanish flint germplasm (Ordás, 1991). The improved synthetics populations EZS1 and EZS2

may be useful for enhancing the genetic basis of breeding programs conducted in similar areas of Europe, and for developing new inbred lines for hybrid production.

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