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Landraces of forage maize as source of genetic variability for organic farming

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Abstract. Concern for the preservation of the environment is a constant at present. The European Commission has developed specific directives to prevent pollution and degradation of the environment. In agriculture, organic farming is presented as an alternative to intensive agriculture that is more polluting and straining on natural resources. Most of the maize breeding materials currently commercialized are not adapted to organic farming conditions and the gene pool has been shrinking after years of genetic selection. Therefore, the landraces used in breeding programs could introduce new genes to obtain materials adapted to new agriculture systems. The aim of this work was to evaluate forage maize hybrids developed at Centro de Investigaciones Agrarias de Mabegondo (CIAM) under Organic Farming. These hybrids were obtained by crossing different Galician landraces with an inbred line which has Galician landrace background. These hybrids have shown good levels of genetic diversity of all studied traits. They were more vigorous and earlier than commercial ones. They had variability of fiber and lignin content but similar digestibility of *in vitro* organic matter, thus breeding programs could discern and exploit these traits. Therefore, landraces could provide new genes to current breeding programs in order to obtain materials more suited to new agricultural conditions.

Keywords. Forage maize – Hybrids – Landraces – Organic Farming.

Les variétés locales de maïs fourrager en tant que source de variabilité génétique pour l'agriculture écologique

Résumé. Le souci de la préservation de l'environnement est actuellement une constante. La Commission européenne a développé des directives spécifiques pour la prévention de la pollution et la dégradation de l'environnement. Dans le domaine de l'agriculture, l'agriculture écologique se présente comme une alternative à l'agriculture intensive, plus polluante et consommatrice de ressources. La plupart des matériaux d'amélioration du maïs actuellement commercialisés ne sont pas adaptés aux conditions de l'agriculture écologique. Le pool génétique a été raccourci après des années de sélection génétique. Par conséquent, l'usage de variétés locales dans programmes d'amélioration pourrait introduire de nouveaux gènes pour obtenir des matériaux adaptés aux nouveaux systèmes agricoles. Le but de ce travail a été d'évaluer les hybrides du maïs fourrager développés au Centro de Investigaciones Agrarias de Mabegondo (CIAM) sous conditions d'agriculture écologique. Ces hybrides ont été obtenus par croisement de différentes variétés locales galiciennes avec une lignée pure, qui possède un fonds génétique de variétés galiciennes. Ces hybrides ont présenté de bons niveaux de diversité génétique pour tous les caractères étudiés. Ils étaient plus vigoureux et plus précoces que les hybrides commerciaux. Ils ont montré une variabilité dans le contenu de fibre et lignine mais la digestibilité *in vitro* de matière organique a été similaire pour tous, ainsi donc les programmes d'amélioration génétique pourraient séparer les génotypes avec meilleurs résultats pour ces caractères. Par conséquent, les variétés locales pourraient fournir de nouveaux gènes aux programmes d'amélioration en cours pour l'obtention de matériaux plus adaptés aux nouvelles conditions de l'agriculture.

Mots-clés. Maïs fourrager – Hybrides – Variétés locales – Agriculture écologique.

I – Introduction

Forage maize is one of the most important sources of roughage for animal feed in Europe, thus its production area exceeded 5 million hectares in 2009 (Eurostat, 2012). The Northwest of Spain concentrates 72% of the global Spanish production which represents 2,500 Megatons of fresh matter (MARM, 2011). During the 80s, landraces were disappearing and were replaced by maize selected hybrids due to their economic advantages. Currently, most of the cultivated maize is hybrid materials from breeding programs, this breeding process being characterized by a shrinking of the genetic background due to the limited number of materials used as parental lines (Barrière *et al.*, 2004).

The increase of organic farming importance in the last decades is causing the demand of new materials adapted to these low input conditions of cultivation. The aim of this work was to evaluate the ability of new hybrids improved from landraces for organic farming use. The narrow genetic background of breeding materials could not be sufficient for obtaining such new materials; moreover the characteristics required for organic or low input farming might have disappeared by selection under high input conditions. Thus a broadening of the genetic base of maize breeding through introduction of new germplasm is necessary. Landraces appear as a good source of new genes and they are earlier, more vigorous, more adapted to environmental conditions and richer in variability making them very useful for breeding programs.

II – Materials and methods

Four maize hybrids were evaluated for agronomic and forage traits under organic farming conditions. During 2008 crosses between inbred line EC49A and landraces 85022, 85042, 85020 and 85040 collected at different Galician sites were made to obtain maize hybrids. Line EC49A was previously obtained at CIAM's breeding programs and has Galician landraces background. Commercial hybrids Nkthermo and Anjou290 (official testers at Spanish Office of Vegetable Varieties - OEVV) were sown too as control.

Field trials were conducted in two years (2009-2010) at four locations: Sobrado (43°0'N, 7°5'W), Baralla (42°5'N, 7°2'W), Pacio (42°6'N, 7°5'W) and Eirexafeita (42°4'N, 7°4'W). The selection of locations was made according to forage maize demand. Experimental design was a randomized block with 8 m² plots and three replications and the final plant density was 90,000 plants per hectare. The agronomic traits that were evaluated were early crop vigour (EV) (subjective ascending scale from 1 to 5) and flowering time (FT) (days from sowing to tasseling). At the silage stage entire plants per plot were cut, weighted and chopped. Samples of 300 g were taken and dried in a ventilated oven at 80°C over the course of 16 hours for the determinations of dry matter yield (DM) and then dried samples were ground in a Christy Norris 8 mill (mesh size= 1mm). Forage quality was determined by near infrared reflectance spectroscopy (NIRS) according to Campo *et al.* (2010) equations. Estimated parameters were *in vitro* organic matter digestibility (IVOMD), crude protein content (CP), acid detergent fiber content (ADF), neutral detergent fiber content (NDF) and lignin content (LIG).

Four experimental environments were taken into account and combined analysis was performed using PROC GLM of the SAS statistical package (SAS, 2008). Hybrids were considered as fixed effects and the Duncan's multiple range test of the LSMEANS statement was used to generate differences among least square means.

III – Results and discussion

Statistical significance of the main effects and means of agronomic, yield and quality traits evaluated in hybrids and controls are shown in Tables 1 and 2, respectively. Highly significant environmental effects were found for all evaluated traits, thus there was an important environmental compound in the observed differences. This shows the different productive ability of the evaluation environments. Different studies have shown the important impact of environmental conditions on the plant fiber content and forage quality, so temperature or radiation exerts stronger impact than the genotype does (Kruse *et al.*, 2008). Genotype effects were also highly significant for evaluated traits (except for DM, IVOMD and LIG). Environment*Genotype interaction was not statistically significant for all traits except for CP, thus the differences between genotypes had remained stable through out the environments so the best genotypes in an environment will be the best on another one.

Hybrids were more vigorous than controls (Table 2). Early crop vigour is associated with higher competitive ability (Pester *et al.*, 1999), so in this way they had better ability to compete with weeds which ensures a better development. This is an important aspect in organic farming where use of inorganic herbicides is not allowed. Among all of the hybrids, 85040 x EC49A and 85020 x EC49A were the most vigorous, not to mention that hybrids were also earlier than controls (74 days vs. 76.6 days), 85022 x EC49A being the earliest within its group. The average DM of hybrids' group was 12.71 t/ha which is slightly higher than the control group, although no significant differences were found between these two groups. Hence, landracexEC49A hybrids could be as productive as commercial hybrids.

Table 1. Statistical significance of main effects of hybrids and controls evaluated at four environments

SV†	EV	FT (days)	DM (t/ha)	IVOMD (%)	CP (%)	ADF (%)	NDF (%)	LIG (%)
Environment (E)	*	***	***	***	***	***	***	***
Rep	*	*	ns	**	**	ns	ns	ns
Genotype (G)	***	***	ns	ns	**	*	**	ns
E*G	ns	ns	ns	ns	**	ns	ns	ns
CV	14.6	2.5	16.3	3.9	10.0	7.6	6.5	9.4

† SV: source of variance; rep: replicate within environments; CV: coefficient of variation.

* P<0.05, ** P<0.01, *** P<0.001, ns: non significant.

Table 2. Means of agronomic, yield and quality traits of hybrids and controls evaluated at four environments

Means	EV	FT (days)	DM (t/ha)	IVOMD (%)	CP (%)	ADF (%)	NDF (%)	LIG (%)
85020 x EC49A	4.58ab†	73.92b	13.69a	65.83b	4.82b	27.55a	53.98a	2.56a
85022 x EC49A	4.08bc	70.92c	11.12b	65.46b	5.10ab	25.46b	50.76bc	2.39bcd
85040 x EC49A	4.33ab	77.83a	12.93ab	63.79b	4.81b	27.48a	54.05a	2.57ab
85042 x EC49A	4.63a	73.68b	12.83ab	66.12b	5.01b	26.12ab	51.70ab	2.50abc
Anjou 290	3.34c	75.16b	12.05b	67.93a	5.46a	24.36b	48.41c	2.26d
Nkthermo	2.50d	78.00a	12.21ab	66.19b	5.56a	25.95ab	50.21bc	2.38cd
Means by group								
Hybrids	4.40	74.06b	12.71	65.29	4.94	26.54	52.49	2.50
Controls	2.92	76.58a	12.13	67.06	5.51	25.16	49.31	2.32
	***	**	ns	*	***	*	**	**

† Means with the same letter were not statistically different.

* P<0.05, ** P<0.01, *** P<0.001, ns: non significant.

The average CP content used to be nearly 8% in silage maize (Barrière *et al.*, 1997). The observed significant reduction in CP content is in agreement with the negative correlation between CP and nitrogen availability found by other authors under low input nitrogen conditions (Lawrence *et al.*, 2008; Li *et al.*, 2010). Lignin content is the primary determinant in cell wall digestibility and hence in organic matter digestibility (Riboulet *et al.*, 2008). Lignin and fiber content were higher in the hybrids group than in control group, although digestibility of the hybrids group was slightly less than that of the control group. Relationship between ADF and NDF is an important trait because it is correlated with NDF digestibility. Thus high feeding value hybrids should have a low ADF/NDF content (Riboulet *et al.*, 2008). ADF/NDF ratio was the same in the hybrids as in control group (ADF/NDF = 0.51). Lignification pattern and biochemical composition of lignin have also an important effect on digestibility, minor variations of these traits could cause an important effect on fiber digestibility, although not on fiber content (Moore and Hatfield, 1994). Despite lignin and fiber content showing great variability among hybrids, IVOMD had similar values and only Anjou 290 showed a significantly different digestibility percentage. The rest of the hybrids had percentages similar to NKthermo control.

IV – Conclusions

Among studied hybrids genetic variability has been observed for all characters except for DM. Landrace x EC49A were as productive as controls for DM but in average they showed better performance of agronomic traits. Of all the hybrids, 85022 x EC49A was the best for FT and quality traits, had the best CP content and had similar values of ADF and NDF to Anjou 290. 85042 x EC49A showed interesting characteristics, having the best EV and values of ADF, NDF and LIG similar to those of NKthermo.

Landrace hybrids proved themselves to be as good as controls; hence a selection program could separate the best genotypes to obtain parental lines from these landraces. Crosses between current inbred lines and these parental lines could very well provide new hybrids better fitted for new agricultural requirements.

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