Identification of teosinte populations (*Zea* spp.) useful for grain yielding improvement in maize (*Zea mays* L.)

Abraham Guerrero-Corona, Lino De la Cruz-Larios*, José de Jesús Sánchez-González, José Ron-Parra, José Alberto Sánchez-Nuño, Eduardo Rodríguez-Guzmán, Fernando Santacruz-Ruvalcaba.

*Instituto para el Manejo y Aprovechamiento de los Recursos Fitogenéticos, Centro Universitario de Ciencias Biológicas y Agropecuarias, Universidad de Guadalajara, km 15.5 carretera Guadalajara-Nogales, Las Agujas, Zapopan, Jalisco, México P. C. 45200

* Corresponding author: E-mail: linocucba@hotmail.com

Keywords: *exotic germplasm, phenotypic, wild relatives.*

Abstract

The great phenotypic and genotypic diversity of Genus *Zea* can be inherited within and between populations. Teosinte (*Zea* spp.) is the closest wild relative to maize, distributed in Mexico and Central America from Chihuahua to Costa Rica through several environmental conditions. The potential ability of exotic germplasm to incorporate traits on maize (*Zea mays* L.) domesticated crops has been demonstrated. Among traits of economic interest that can be transferred from teosinte to maize, the following stand out: higher grain yield, resistance to pests and diseases, and product quality. 180 crosses between BC_2F_1 (maize-teosinte families) and LUG282 were evaluated to test introgressed teosinte germplasm potential on CIMMYT line CML311 background. The 180 F_1 with teosinte introgressions were evaluated at three environments and compared to a reference control LUG282xCML311 and to some other experimental and some commercial hybrids as controls also. Main variables evaluated were days to anthesis and silking, plant and ear height, root and stalk lodging and grain yield. The results of the combined ANOVA by teosinte families showed that hybrids with introgressions of a teosinte population from La Lima, Tolimán, Jalisco, (*Zea mays* ssp. *parviglumis* landrace Balsas) averaged higher in grain yield, but they were not statistically superior to the reference control LUG282xCML311 ($\alpha = 0.05$); while in the combined ANOVA by treatments only the hybrid with teosinte T100 (T = treatment number) was statistically superior to reference control for grain yield ($\alpha = 0.05$). Among other traits, hybrids with *Zea diploperennis* introgressions (San Andres Milpillas, Nayarit) appeared to be a reliable source for resistance to foliar diseases.

Abbreviations

DTA – days to anthesis
DTS – days to silking
PH – plant height
EH – ear height
NP – number of plants
SL – stalk lodging
RL – root lodging
EN – ear number;
EW – ear weight
GW – grain weight.

Introduction

Maize (*Zea mays* L.) is a plant cultivated around the world for its great importance in human and livestock nutrition, and for being used widely in the industry, in products such as starches, oils, varnishes, paints, plastic and soaps, among others (Song *et al.*, 2012). In Mexico, it is crucial to continue producing both high-yielding and high-quality hybrid varieties of white maize for direct human consumption, and yellow maize for the processed food industry. To achieve this goal, it is important to explore new germplasm sources with potential to expand the populations currently used in breeding programs.

Teosinte species grouped in the genus *Zea* (Doebley, 1990; Iltis and Benz, 2000; Gomez-Laurito, 2013) represent an important source of exotic germplasm as they grow in a wide variety of ecological conditions, from warm and humid regions in the south of Mexico and Central America to cold and dry valleys in northern and central Mexico. Teosinte can be found on edges and within maize fields, on small stream banks, in open forests on rocky mountain slopes, and in grassland areas as an herbaceous cover constituent.

The high adaptation of teosinte to several special ecological conditions represents a great potential for discovering new traits, not present in modern maize. For example, *Zea nicaraguensis* carries flood resistance genes (Mano and Omori, 2007; Mano *et al.*, 2008; Mano *et al.*, 2013; and Mano and Omori, 2015); and *Zea luxurians* from San Felipe Usila, Oaxaca, where it rains about 4000 mm annually, may also be a source of flood resistance alleles. On the other hand, in the dry environments of Durango valleys, *Zea mays* ssp. *mexicana* seems to survive due to a noticeably short period of vegetative growth and drought resistance genes (Sanchez *et al.*, 2018). Nault, (1982) found that *Zea perennis* and *Zea diploperennis* possess resistance against various viruses attacking maize. Further, one of the few resistance species to *Striga* spp., a parasitic plant of maize roots is *Zea diploperennis* (Rich and Ejeta, 2008). Moreover, Flint-García and Bodnar, (2009) evaluated the chemical characteristics of maize and teosinte kernels and found that the latter has smaller seeds than maize but twice its protein content. The potential use of teosinte in maize breeding has been evaluated since the 1950s and several researchers concluded that teosinte may be a valuable germplasm for maize improvement (Reeves, 1950; Sehgal, 1963; Cohen and Galinat, 1984, Casas *et al.*, 2003). Additionally, it has been verified that teosinte germplasm can be incorporated to maize and persists in advanced generations of backcrossing (Rincón, 2001; Kato and Sánchez, 2002). Wang *et al.*, (2008) incorporated *Zea mays* ssp. *mexicana* germplasm to Ye515 maize elite line. After two backcrosses and four cycles of selfing, recovered lines showed great variation in ear characteristics, resistance to various diseases and chemical composition of grain. The backcrossing method is useful to make teosinte alleles more readily available for using in maize breeding programs. For these reasons, we developed BC_2F_1 families and crossed by a single inbred line obtaining hybrids with predominant maize genetic backgrounds and produce essentially normal maize plants and ear phenotypes carrying small proportions of unique teosinte introgressions.

The aim of this research was to identify teosinte populations with potential to improve grain yield, flowering time, and stalk strength, among other traits that may be useful in maize germplasm.

Material and methods

Germplasm

The original population was provided by the Germplasm Bank of Centro Universitario de Ciencias Biológicas y Agropecuarias (CUCBA). From this reference population with 900 BC_2F_1 maize-teosinte families, 180 were selected, of which every group of 9 families carries introgressions from a single teosinte population (it is important to highlight that one plant per population was used as a founder to form F1), therefore, 20 original populations were represented in this sample of 180 families including the following species and subspecies: two populations of *Zea diploperennis* (Iltis, Doebley & Guzmán) from states of Jalisco and Nayarit, two populations of *Zea luxurians* (Durieu & Ascherson) Bird, from Oaxaca and Guatemala, 6 populations of *Zea mays* ssp. *mexicana* (Schrad.) Iltis (landraces Chalco, Central Plateau, Durango and Nobogame), 10 populations of *Zea mays* ssp. *parviglumis* (Iltis & Doebley) landrace Balsas, and one population of *Zea mays* ssp. *huehuetenangensis* (Iltis & Doebley) Doebley from Guatemala. (More details of populations origin on Table S2). BC_2F_1 genetic background is composed as follows: 87.5 % from recurrent parent (CIMMYT inbred line CML311) and 12.5 % from donor parents (species and subspecies of teosinte) representing a wide distribution and diversity of species and subspecies of teosinte from Mexico and Guatemala (Fig. 1).

Fig. 1a - Map of Mexico and Central America with records of teosinte populations.

Hybrid development

To form hybrids and their subsequent evaluation, the inbred line LUG282 was used as female parent, and 180 BC2F1 families as male parents; These crosses were made during the 2014 summer cycle and the 2014-2015 winter cycle at CUCBA Experimental Agricultural Field. Once F1 (LUG282xBC2F1) crosses were harvested in both cycles, evaluation trials were carried out in the 2015 summer cycle.

Phenotypic evaluation trials

During 2015, trials were sown in June, under rainfall summer season conditions at three different environments in Jalisco, Mexico: La Soledad, Zapotlan del Rey, with annual mean temperature 20.1 °C, annual mean rainfall 819 mm and altitude 1530 m; El Salitre, San Martin de Hidalgo, with annual mean temperature 20.9 °C, annual mean rainfall 964 mm and altitude 1260 m; and CUCBA experimental field, Zapopan, with annual mean temperature 18°C, annual mean rainfall 816 mm and altitude 1650 m. The 180 hybrids were compared with the reference control hybrid LUG282xCML311 (without teosinte introgressions). Another two experimental hybrid controls used were LUG78xCML311, LUG03xCML311, and five commercial hybrids from private companies: DK2027Y, Cimarron, P3055W, DAS2362 and P3164W.

Experimental design

Experimental design on locations was alpha-lattice 12x16, composed by 192 hybrids (180 maize-teosinte hybrids plus 12 controls) on 16 incomplete blocks, with 12 hybrids per block and three replications), resulting in 576 plots per location. Design development and randomization were carried out using Plant Breeding tools (PBtools) software from International Research Rice Institute (IRRI). The plot size was 2 rows of 4 m long each with 0.76 m distance between rows and 1 m alleys between blocks.

Agronomic management

Trial planting was carried out manually at localities La Soledad and El Salitre, while in CUCBA Almaco brand experimental seeder was used. Conventional agronomic work was carried out in accordance with specific maize crop management schemes for each locality, for planting, fertilization, monitoring and timely control of pests and weeds. Harvesting and recording of variables were carried out manually in El Salitre and CUCBA Experimental Agricultural Field, while in La Soledad the trial was harvested with a New Holland Twin Rotor 88 experimental harvesting machine, at these trial harvest variables obtained were:grain moisture, grain weight per plot; grain yield was adjusted to kilograms per hectare (kg/ha).

Measured variables

Variables measured per plot in every trial were as follows:

a) Phenological: days to anthesis (DTA), consisted of counting the number of days when 50% of plants in plot initiated pollen shed; days to silking (DTS), recorded when 50% of plants exposed maize shoots with stigmas longer than 3 cm;

b)Agronomical: plant height (PH), measured from the stalk base to insertion of banner leaf, and ear height (EH), measured from the stalk base to the node where the ear rachis emerges, both registered in centimeters; number of plants (NP), counted at the end of flowering stage to determine population densities; stalk lodging (SL), number of broken or folded stalk plants counted one week before harvest; root lodging (RL), number of plants with inclined stalks in an angle of more than 45°; ear number (EN) per plot; ear weight (EW), total ear weight in kg; grain weight (GW), per plot adjusted to 14% moisture. To adjust grain weight (GW) to 14 % moisture in El Salitre and CUCBA trials, ears per plot were mechanically shelled, grain was weighted in kg and weight adjusted. At La Soledad the machine harvester recorded grain weight, and grain yield per plot was adjusted to kg/ha.

Data analysis

Two combined analysis of variance with three trial locations (environments) were performed using procedure GLM from SAS (SAS Institute, 2013), by treatment and by treatments nested in original teosinte family. Dunnett's test (α = 0.05) was performed to compare least square means for grain yield and agronomic characteristics between reference control and hybrids with teosinte introgressions. The performed mixed linear model by treatment (hybrid) was as follows:

Response Variable = μ + Location + Replication (Location) + Treatment + Location*Treatment + Location*Replication (Location)

The performed mixed linear model by teosinte family was as follows:

Response Variable= μ + Location + Replication (location) + Teosinte family + Location*Teosinte family + Location*Replication (Location)

The interactions Location*Replication (Location) were considered random effects.

Results

The combined ANOVA by treatment showed highly significant differences (α = 0.01) for most phenotypic traits in variation sources: Locations, Replications (Locations) and Treatments. For interaction Locations by Treatments there were not significant differences in ear height and root lodging (Table 1). This response points out to a strong interaction within genotypes (Treatments) and between the contrasting environments of Locations. The overall time interval between days to anthesis and days to silking was 0.86 d. Observing the common protandry in maize, there are marked differences in cycle between hybrids with introgressions from northern and southern teosinte populations, being northern hybrids earlier than southern ones. Regarding plant and ear height teosinte germplasm seemed to increase height average compared to reference

Table 1 - Combined analysis of variance from three environments: (La Soledad, El Salitre and CUCBA)

Source	D. F.	DTA	DTS	PH	EH	RL	SL	GW	Yield/ha	D. F.	EN
Location	2	3726.90**	3319.95**	269078.69**	258329.59**	1346.72**	3012.09**	935.89**	2212301026**	1	6118.28**
Replication (Loc)	5	57.28**	40.64**	4126.17**	1052.63**	2050.16**	18.15**	12.64**	24807814**	3	960.50**
Treatments	187	10.63**	9.51**	526.18**	470.80**	93.78**	37.94**	3.53**	7192077**	185	72.45**
Loc*Treat	368	6.40**	5.80**	189.84**	114.75 ns	58.35 ns	28.92**	1.14**	2448278**	183	50.64**
M. S. E.		3.94	3.36	146.67	107.66	83.14	25.88	0.88	1981758		41.53
Coeff. Var.		2.94	2.68	4.78	7.36	105.84	107.46	15.02	15.54		14.93
Mean		67.60 d	68.46 d	253.23 cm	141.05 cm	8.61 %	4.73 %	6.25 kg/pl	9057.36 kg/ha		43.15 ears

**= Highly significant to 0.01, *= Significant to 0.05, ns = no significant. D. F. = Degrees of Freedom, M. S. E. = Mean Square Error, C. V. = Coefficient of Variation. Traits, DTA = Days to Anthesis (d), DTS = Days to Silking (d), PH= Plant Height (cm), EH = Ear Height (cm), RL= Root Lodging percentage (%), SL = Stalk Lodging percentage (%),GW= Grain Weight (kg/plot), Yield/ha = Adjusted Yielding (kg/ha), EN = Ear Number (count).

control, which made hybrids susceptible to root and stalk lodging. Coefficient of variation for main variables grain weight and adjusted yielding to kg/ha were 15.02 % and 15.54 % respectively, acceptable values for this type of experiments in which environmental conditions influence plant germplasm performance and plasticity. Another important trait measured in two locations (El Salitre and CUCBA) was ear number, an increment in prolificacy was observed in hybrids with teosinte introgressions.The combined ANOVA by teosinte family showed great interaction between environments

and treatments when grouped by their original teosinte donor parent, except for root lodging, where there were not significant differences (Table 2). Regarding other traits, there were highly significant differences (α = 0.01) in every source of variation, despite a small introgression of 6.25 % in the hybrids genetic background; this may be attributable to the diverse teosinte introgressions from 20 populations involved in these germplasms and expressed in the phenotypes of 180 hybrids through locations.

Dunnett's test (Table S1) showed treatments that were significantly different (LSD 0.05) compared to reference control LUG282xCML311. For both variables DTA and DTS there were 12 treatments with teosinte germplasm that were earlier than reference control (67.16/68.73 d): T2 64/65.11 d and T3 65.16/66.44 d (introgression from population San Andres Milpillas, *Zea diploperennis*), T30 64.11/64.94 d (El Pedregal, ssp. *mexicana* landrace Chalco), T57 64.72/65.94 d (ssp. *mexicana* landrace Central Plateau), T144, T135, T147 and T150 (ssp. *parviglumis* landrace Balsas) with 65.27/65.66 d,

Table 2 - Combined analysis of variance by teosinte groups used as donors.

Source	D. F.	DTA	DTS	PH	EH	RL	SL	GW	Yield/ha	D. F.	EN
Location	2	2542.79**	1245.75**	265025**	244450.61**	409.63**	2112.39**	529.69**	1231700898**	1	1890.33**
Replication (Loc)	5	89.57**	63.91**	4937.19**	858.87**	2050.16**	18.15**	12.64**	24807814**	3	960.50**
Teosinte family	27	14.45**	15.84**	1045.50**	1305.23**	202.33**	41.86**	12.47**	25149184**	27	137.69**
Loc*Teo	54	7.32**	5.87**	231.27**	113.86**	41.58 ns	43.15**	1.88**	3794944**	27	51.80**
M. S. E.		6.29	5.15	202.01	130.35	79.69	27.55	1.05	2299035		47.13
Coeff. Var.		3.68	3.26	5.64	7.95	103.62	110.88	16.38	16.74		15.91
Mean		68.23 d	69.65 d	252.06 cm	143.65 cm	8.61 %	4.73 %	6.25 kg/pl	9057.36 kg/ha		43.15 ears

**= Highly significant to 0.01, *= Significant to 0.05, ns= no significant. D. F.= Degrees of Freedom, M. S. E.= Mean Square Error, C. V.= Coefficient of Variation. Traits: DTA= Days to Anthesis (d), DTS= Days to Silking (d), PH= Plant Height (cm), EH= Ear Height (cm), RL= Root Lodging percentage (%), SL= Stalk Lodging percentage (%), GW= Grain Weight (kg/plot), Yield/ha= Adjusted Yielding (kg/ha), EN= Ear Number (count).

Table 3 - Dunnett's T test for teosinte families least squares means for traits evaluated through three environments.

*Teosinte source	DTA	DTS	PH	EH	RL	SL	EN	Yield/ha
1 San Andres Milpillas (*Zea diploperennis*)	66.70**	67.84**	237.41**	124.39**	6.38**	4.64**	38.58	8009.38**
2 Tarahumares (ssp. *mexicana landrace* Nobogame)	66.82**	68.11**	244.66**	133.87**	5.57**	4.69**	41.49**	8670.63**
3 Potrero El Tepalcate (ssp. *mexicana landrace* Durango)	66.86	68.09**	252.08	140.97**	9.50	5.56**	43.91**	8561.22**
4 El Pedregal (ssp. *mexicana landrace* Chalco)	66.58**	67.58**	247.63**	132.18**	8.30	4.23**	39.18	8165.09**
5 Opopeo (ssp. *mexicana landrace* Chalco)	66.79**	67.86**	248.47	134.78**	8.68	4.55**	40.61	8566.35**
6 El Salteador (ssp. *mexicana landrace* Central Plateau)	67.20	68.17**	248.21	138.16	5.65**	3.81**	42.24**	8091.96**
7 Penjamillo de Degollado (ssp. *mexicana landrace* Central Plateau)	67.33	68.09**	249.14	137.82	7.55	5.00**	41.95**	9009.26**
8 Camino Carboneras (ssp. *parviglumis landrace* Balsas)	67.36	68.48	245.90**	135.31**	8.19	4.76**	42.39**	8415.74**
9 San Miguel Cuzalapa (*Zea diploperennis*)	67.24	68.42	246.03**	132.81**	6.04**	5.04**	42.51**	8545.99**
10 Crucero Lagunitas (ssp. *parviglumis landrace* Balsas)	67.46	68.47	258.62**	147.90**	9.99**	5.88	46.88**	9074.21**
11 Los Cimientos (ssp. *parviglumis landrace* Balsas)	68.41**	69.50**	256.59**	145.42**	11.63**	7.72	45.70**	8929.96**
12 El Salado (ssp. *parviglumis landrace* Balsas)	67.70	68.94	256.97**	145.53**	12.47**	5.05**	42.76**	8909.64**
13 San Cristobal Honduras (ssp. *parviglumis landrace* Balsas)	67.39	68.26**	247.24**	134.16**	6.96	4.90**	42.55**	8280.23**
14 Huixtitla (ssp. *parviglumis landrace* Balsas)	67.12	67.99**	251.10	142.81**	8.95	6.46	43.10**	8649.50**
15 La Lima (ssp. *parviglumis landrace* Balsas)	67.40	68.22**	253.00**	140.22**	8.42	6.44	42.66**	9253.72**
16 Zacatlancillo (ssp. *parviglumis landrace* Balsas)	66.94	67.85**	251.53	140.74**	9.82	3.94**	45.31**	9113.25**
17 El Potrero (ssp. *parviglumis landrace* Balsas)	67.74**	68.67	249.26	141.65**	9.39	4.57**	43.34**	8605.31**
18 Plan de los Timbres (ssp. *parviglumis andrace* Balsas)	67.87**	68.64	254.15**	143.10**	11.69**	5.68**	44.46**	9110.33**

*Teosinte source	DTA	DTS	PH	EH	RL	SL	EN	Yield/ha
19 Las Majadas (Zea luxurians)	68.31**	69.38**	248.06**	136.19**	6.80	3.33**	43.73**	8118.06**
20 El Tablon (ssp. huhuetenangensis)	67.80**	68.96	256.79**	143.29**	10.97**	5.45**	41.65**	8733.78**
21 LUG282xCML311	67.16	68.73	250.17	137.92	8.17	6.73	39.82	10080.06
22 CML78xCML311	66.89	68.33	232.01**	129.74**	1.33**	3.10**	42.58**	9473.42**
23 LUG03xCML311	67.56	69.17	229.46**	129.93**	0.66**	9.90**	42.5**	10243.06
24 DK2027Y	67.44	68**	249.00	132.54**	1.17**	2.25**	41.42	11620.96**
25 Cimarron	67.72	67.83**	242.06**	125.02**	0.33**	4.67**	41.25	12403.40**
26 P3055W	64.66**	65.55**	260.06**	136.22	0.52**	2.69**	43.66**	12251.32**
27 DAS2362	67.39	68.61	249.80	129.48**	1.19**	3.68**	36.16**	10337.21
28 P3164W	64.72**	65.16**	234.01**	122.37**	0.55**	6.08	43.25**	12496.33**

** Minimal significant differences to 0.05 probability. Traits: DTA= Days to Anthesis (d), DTS= Days to Silking (d), PH= Plant Height (cm), EH= Ear Height (cm), RL= Root Lodging percentage (%), SL= Stalk Lodging percentage (%), GW= Grain Weight (kg/plot), Yield/ha= Adjusted Yielding (kg/ha), EN= Ear Number (count).

65.33/66.16 d, 65.72/66.72 d and 65.72/67.05 d, respectively, T26 with 65.33/66.38 d (Potrero El Tepalcate, ssp. *mexicana* landrace Durango) and T40, T42 and T39 with 65.38/67.22 d, 65.50/66.50 d and 65.66/66.83 d, respectively (Opopeo, ssp. *mexicana* landrace Chalco). The commercial hybrids P3055W and P3164W were earlier than reference control with 64.66/65.55 d and 64.72/65.16 d respectively. While the significant later treatments were 8: T98 with 70.22/71.24 d (ssp. *parviglumis* landrace Balsas) and T179 with 70.27/71.16 d (ssp. *huehuetenangensis*) were the latest. For traits PH and EH the shortest height treatments with teosinte germplasm were T2, T9, T3 and T8 with 229.94/114.91 cm, 231.10/124.43 cm, 232.40/113.51 cm, and 233.22/122.77 cm, being the teosinte family from San Andres Milpillas, Nayarit the most prevalent (*Zea diploperennis*). Plants with tall archetype were treatments with germplasms of *Zea mays* ssp. *parviglumis* landrace Balsas (T144, T160, T84, T96, T141, T90, T92, T127, T97, T100, T99, T86, T88 and T133) and *Zea mays* ssp. *huehuetenangensis* (T176, T178, T177 and T173). Regarding RL and SL variables there were meteorological conditions that favored lodging due to uncommonly high-speed winds (Patricia hurricane was present that year), being treatments with germplasm of *Zea mays* ssp. *parviglumis* landrace Balsas the most susceptible due to their tallest plants. Root lodging ranged between 21.78 % and 16.41 % while stalk lodging ranged 17.73 % and 11.31 %.

For main trait adjusted grain yielding to kg/ha only treatment T100 (El Salado, ssp. *parviglumis* landrace Balsas) with teosinte germplasm was significantly superior (α = 0.05) to reference control, commercial hybrid DAS2362 and experimental hybrids CML78xCML311 and LUG03xCML311; showing that, despite the great diversity of teosinte germplasm only some populations from the teosinte lowlands *Zea mays* ssp. *parviglumis* brought an increase in grain yield. No treatment with teosinte was superior to the other commercial hybrids P3055W, P3164W, DK2027Y and Cimarron. EN trait demonstrated that teosinte germplasm introgressed in maize background increases ear prolificacy, there were 40 significant prolific treatments being the best ones: T99, T84, T96 and T146 with 53.16, 51.58, 50.25 and 50.1 ears per plot respectively, and ssp. *parviglumis* landrace Balsas and T164 (*Zea luxurians*) with 50.4 ears per plot.

In Dunnett's test performed by families of teosinte sources compared to controls, treatments grouped by families did not exceed neither reference control (LUG282xCML311) nor other commercial hybrids, and the best teosinte family for grain yield was *Zea mays* ssp. *parviglumis* landrace Balsas from La Lima, Jalisco (Table 3). Population 1 germplasm from the state of Nayarit *Zea diploperennis* and teosintes from central highlands of Mexico *Zea mays* ssp. *mexicana* landrace Chalco brought flowering time earlier. Population 1 (*Zea diploperennis*, Nayarit) and population 2 (ssp. *mexicana* landrace Nobogame, from the state of Chihuahua) brought shorter plants; and population 10

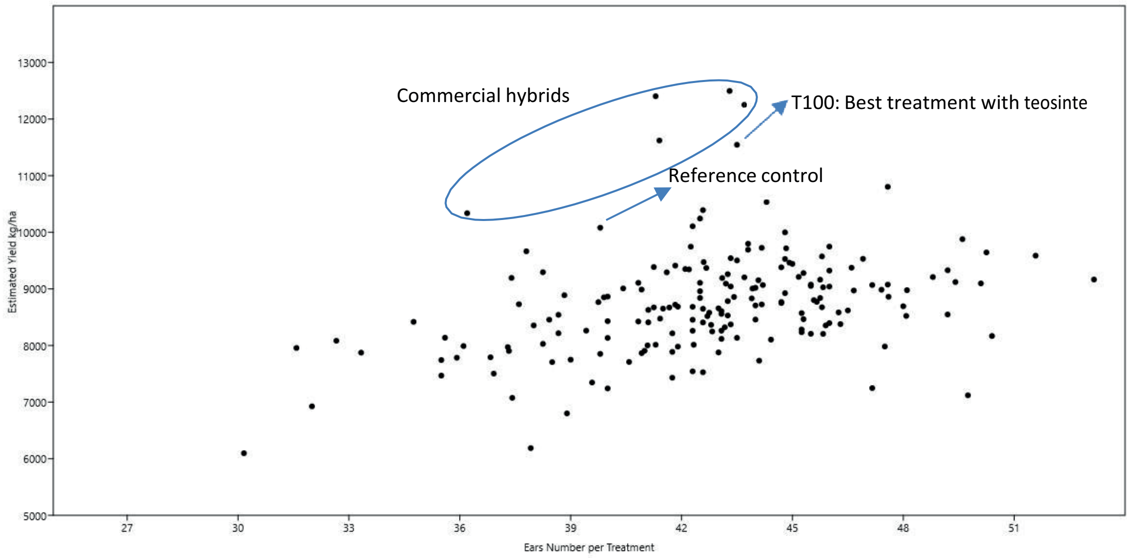

Fig. 2 - Dispersion diagram of grain yield and ear number from evaluation of the 180 treatments of maize hybrids with teosinte introgressions plus 12 controls of maize hybrids.

appeared to be the best in terms of prolificacy (*Zea mays* ssp. *parviglumis* landrace Balsas, Guerrero).

Most hybrids with incorporated teosinte germplasm tend to increment ear number per plant, a trait called prolificacy (Figure 2), but not necessarily an increase in grain weight, compared to reference control and commercial hybrids. The reference control has a ratio of 0.90 ears per plant, the best yielding hybrid with teosinte germplasm has 0.95 ears per plant (T100, *Zea mays* ssp. *parviglumis* from El Salado, Mochitlán Guerrero), and commercial hybrids possess mean ratio of 0.90 ears per plant. The most prolific treatment was T164, a hybrid with germplasm of *Zea luxurians* from Las Majadas, Jutiapa, Guatemala. It can be observed that 70.55 % hybrids with teosinte germplasm (127 treatments) were not superior to the reference control, and 28.33 % of these hybrids (51 treatments) were statistically similar to the reference control, which seems to indicate that despite the wide diversity of genus *Zea*, only a small number of teosinte populations may represent a reliable source for increasing grain yield in maize; nevertheless, genus *Zea* has unique traits that could be usable in the future, such as resistance to pest and diseases, resilience to climatic change, perennial crops, and tolerance to flooding conditions, among others.

Discussion

As stated above, previous studies regarding the use of wild relatives, like different populations of teosinte to improve maize can be found in the literature since the 1950s. It has been done with relative success, improving or expanding traits of economic importance (Reeves and Mangelsdorf, 1959; Lambert and Leng, 1965; Cohen and Galinat, 1984; Casas *et al.* 2003; Padilla *et al.* 2002; Wang *et al.* 2008). Most recent investigations such as Rosas *et al.* (2015) have evaluated phenotypic traits with agronomic functions in maize lines with different levels of introgression (backcrosses two and three) of *Zea mays* ssp. *parviglumis* landrace Balsas, ssp. *mexicana* (landraces Chalco and Central Plateau) and *Zea diploperennis* from Jalisco. Based on the above, this work tried to identify a greater number of teosinte germplasm sources helping to enhance fundamental traits such as grain yield, earliness and resistance to lodging in maize crops, as teosinte species genetic pool counts with unique traits not present in modern maize landraces and improved cultivars. Including larger numbers of populations representing known species, subspecies and landraces diversity, among them: teosinte landraces Central Plateau, Chalco, Durango and Nobogame (*Zea mays* ssp. *mexicana*), and *Zea diploperennis* from Nayarit and Jalisco, from the highlands; and the wide distribution through the southern lowlands: teosinte landrace Balsas (*Zea mays* ssp. *parviglumis*), the populations from Guatemala landrace Huehuetenango (*Zea mays* ssp. *huehuetenangensis*) and the species *Zea luxurians*, is considered of great importance. Each of the previous investigations referred above has the particularity of having worked with a reduced diversity of teosinte species and closely related families; in this research, a structure of introgressed populations (Prohens *et al.* 2017) was implemented to evaluate most of the available and known species of teosinte these days. Incorporating the greatest number of teosinte populations known is justified into a study such as this one given that it seeks to capture genetic variability bringing increments in quantitative traits such as grain yielding of maize elite inbred lines where teosinte germplasm has been incorporated, mainly by backcrossing inbred lines method as suggested by Jeuken and Lindhout (2003).

The assessment carried out here allowed for a broad evaluation of phenotypic traits such as grain yield, flowering time, plant and ear height, and root and stalk lodging, since the 20 BC2F1 CML311-teosinte families crossed by female inbred LUG282 carry a wide variability of introgressions. In this way, evaluating these crosses allowed to directly identify which species of teosinte are involved with significant changes, compared to reference control (without teosinte background) in the measured variables. The best commercial hybrids evaluated for these locations have very good performance, beating the reference control LUG282xCML311 and all crosses with maize-teosinte BC2F1 families. It is important to point out that the twenty teosinte families (experimental hybrids nested by teosinte source) were significantly inferior for grain yield versus commercial hybrid controls and reference control (Table 3). In relation to other agronomic traits considered, there are very few remarkable differences between teosinte crossings compared to reference control. However, there is a trend towards higher plants and ear height similar to that found in previous works such as Rosas *et al.* (2015) and for this reason increased root lodging, especially in hybrids with *Zea mays* ssp. *parviglumis* and *Zea mays* ssp. *huehuetenangensis* introgressions. The lower the genetic variation in breeding populations, the less likely breeders can identify new and useful combinations of genes; at the same time, wild germplasm, an underutilized reservoir of genetic variation can contribute to widen domesticated crop species diversity (Tanksley and Nelson 1996). Incorporating different teosinte species germplasm transferred to CML311 inbred line through backcrossing method has led to an observable variation in the progenies even when families were backcrossed for two rounds only getting 12.5 % of teosinte genetic introgressions on 87.5 % CML311 genetic background, contrasting with other works carried out with teosinte pollen mixtures such as Chuela (1999), Padilla *et al.* (2002) and Casas *et al.* (2003).

According to results obtained from the combined analysis of variance and Dunnett's test, the outstanding teosinte sources for grain yielding are populations from La Lima, municipality of Tolimán, Jalisco; and treatment T100, with introgression from a population located at El Salado, municipality of Mochitlan, Guerrero, both corresponding to *Zea mays* ssp. *parviglumis* introgressions, the teosinte species most closely related to maize (Rivera-Rodriguez *et al.* 2019). For earliness, treatments with *Zea diploperennis* germplasm from San Andres Milpillas, Huajicori, Nayarit were significant, and landraces Nobogame and Chalco (*Zea mays* ssp. *mexicana*) showed a good source of variation for this earlier flowering, while for resistance to foliar diseases *Zea diploperennis* from Jalisco may represent one of the best options. The results previously described suggest that the outstanding BC2F1s are a very important guide to follow our research towards phenotypic evaluations in a greater number of environments, with extensive sampling and a detailed search for favorable alleles in the populations evaluated with individual teosinte plants. It should be highlighted that the 180 BC2F1 families used are a sample of a population of 900 BC2F1, which will be derived in more breeding cycles to BC3F3, with the purpose of evaluating these maize-teosinte isogenic lines; therefore, this is a preliminary work that provides a preview of the potential of developing this new reference population.

References

Casas S JF, Ramírez D JL, Sánchez G JJ, Ron P J, Montes H S, Chuela B M. 2003. Características agronómicas en retrocruzamientos maíz-teocintle. Fitotecnia Mexicana. 26 (4): 239 – 248.

Chuela B M. 1999. Análisis genético de las cruzas dialélicas de seis líneas élite de maíz con germoplasma de teocintle. *Thesis.* M. C. Manejo de áreas de temporal. Centro Universitario de Ciencias Biológicas y Agropecuarias. Universidad de Guadalajara.

Cohen JI, Galinat WC. 1984. Potential Use of Alien Germplasm for Maize Improvement. *Crop Science*. 24: 1011-1015.

Doebley JF. 1990. Molecular systematics of *Zea (Graminae). Maydica*. 35: 143-150.

Flint-Garcia SA, Bodnar AL, Scott MP. 2009. Wide variability in kernel composition, seed characteristics, and zein profiles among diverse maize inbreds, landraces, and teosinte. *Theoretical Applied Genetics*. 119: 1129-1142.

Gomez-Laurito J. 2013. A new species of *Zea* (*Poaceae*) from Murcielago Islands, Santa Elena Peninsula, Guanacaste, Costa Rica. Brenesia. 80: 36-39.

Iltis HH, Benz BF. 2000. *Zea nicaraguensis* (*Poaceae*), a new teosinte from Pacific coastal Nicaragua. Novon. 10: 382-390.

Jeuken M. J., Lindhout P. 2003. Future perspectives of Backcross Inbred Lines for exploitation of wild germplasm: a case study on Lactuca saligna as a donor for quantitative resistance to lettuce downy mildew. Eucarpia Leafy Vegetables. 69-74.

Kato Y TA, Sánchez G JJ. 2002. Introgression of chromosome knobs from *Zea diploperennis* into maize. Maydica. 47: 33-50.

Lambert RJ, Leng ER. 1965. Backcross response of two mature plant traits for certain corn-teosinte hybrids. *Crop Science*. 5: 239-241.

Mano Y, Omori F. 2007. Breeding for flooding tolerant maize using "teosinte" as a germplasm resource. Plant Root. 1: 17-21.

Mano Y, Omori F. 2013. Relationship between constitutive root aerenchyma formation and flooding tolerance in *Zea nicaraguensis*. Plant Soil. 370: 447-460.

Mano Y, Omori F, Loaisiga CH, Bird RM. 2008. QTL mapping of above - ground adventitious roots during flooding in maize x teosinte "*Zea nicaraguensis*" backcross population. Plant Root. 3: 3-9.

Mano Y, Omori F. 2015. Flooding tolerance in maize (*Zea mays* ssp. mays) F1 hybrids containing a QTL introgressed from teosinte (Zea nicaraguensis). Euphytica. 359 205:255–267.

Nault LR, Gordon DT, Iltis HH. 1982. Response of annual and perennial teosintes (*Zea*) to six maize viruses. Plant Disease. 66 (1): 61-62.

Padilla GJM, Sánchez GJJ, Ramírez DJL, Casas SJF, Ron PJ, Chuela BM, Aguilar SM. 2002. Medias y varianzas en variedades sintéticas de maíz con diferentes fuentes de germoplasma de teocintle. Fitotecnia Mexicana 25 (4): 401-409.

Prohens J, Gramazio P, Plazas M, Dempewolf H, Kilian B, Diez MJ, Fita A, Herraiz FJ, Rodriguez-Burruezo A, Soler S, Knapp S, Vilanova S. 2017. Introgressiomics: a new approach for using crop wild relatives in breeding for adaptation to climate change. Euphytica 213:158.

Reeves RG. 1950. The use of teosinte in the improvement of corn inbreds. Agronomy Journal. 42: 248-251.

Reeves RG, Mangelsdorf PC. 1959. The origin of corn. II. Teosinte a hybrid of corn and Tripsacum. Botanical Museum of Leaflets. Narv. Univ. 18:357–387.

Rich PJ, Ejeta G. 2008. Towards effective resistance to Striga in African maize. Plant Signal Behavior. 3(9): 618–621.

Rincón EG. 2001. Analisis molecular del flujo genético teocintle-maíz en México. Tesis de Maestría, Colegio de Postgraduados, Montecillo, México.

Rivera-Rodríguez DM, Sánchez-González JJ, De la Cruz L L, Santacruz-Ruvalcaba F, Ruiz-Corral JA. 2019. Morphological and Climatic Variability of Teosinte (*Zea* spp.) and Relationships Among Taxa. Systematic Botany. 44 (1), 41-51.

Rosas M O, Sánchez-González JJ, De la Cruz L L. 2015. Retrocruza maíz-teocintle potencial de aprovechamiento en el mejoramiento genético del maíz (*Zea mays* L.).

Thesis. M. C. Biosistemática y Manejo de Recursos Naturales y Agrícolas. Centro Universitario de Ciencias Biológicas y Agropecuarias. Universidad de Guadalajara.

Sánchez G JJ, Ruiz C JA, Medina GG, Ramirez GO, De la Cruz LL, Holland JB, Miranda RM, Garcia R GE. 2018. Ecogeography of Teosinte. Plos One. 13: 387 e0192676. SAS Institute. 2013. SAS Version 9.4. The SAS Inst. Inc., Cary, NC.

Sehgal SM. 1963. Effects of teosinte and "Tripsacum" introgression in maize. Ph. D. Dissertation, Bussey Institute, Harvard University, Cambridge, Massachusetts.

Song RT, Wang G, He QQ, Xu ZK. 2012. High segregation distorsion in maize B73 x teosinte crosses. Genetics and Molecular Research. 11 (1): 693-706.

Tanksley SD, Nelson JC. 1996. Advanced backcross QTL analysis: a method for the simultaneous discovery and transfer of valuable QTLs from unadapted germplasm into elite breeding lines. Theoretical and Applied Genetics. 92: 191-203.

Wang L, Yang A, He C, Qu M, Zhang J. 2008. Creation of new maize germplasm using alien introgression from *Zea mays* ssp. *mexicana*. Euphytica. 164:789–801.

Table S1 - Dunnett's T test for least squares means for traits evaluated through three environments.

Treatment	*Teosinte source	DTA	DTS	PH	EH	RL	SL	EN	Yield/ha
T1	1	66.89	67.89	245.86	131.40	6.69	1.33**	34.75	8417.63**
T2	1	64**	65.11**	229.94**	114.91**	0.67	5.67	37.41	7074.96**
T3	1	65.16**	66.44**	232.4**	113.51**	1.56	3.14	41.08	8001.86**
T4	1	67.06	68.78	238.47**	128.86**	9.20	4.82	32.66**	8083.70**
T5	1	67.00	67.83	242.38	121.68**	13.12	2.19	41.25	8672.80**
T6	1	66.78	68.00	239.36**	129.96**	7.77	6.81	40.83	8424.32**
T7	1	67.61	68.50	244.03	131.98	10.77	3.41	38.5	7706.31**
T8	1	68.94**	69.56	233.22**	122.77**	6.01	8.22	35.5	7468.26**
T9	1	66.89	68.50	231.1**	124.43**	1.67	6.26	45.25	8234.60**
T10	2	66.56	68.56	243.21	129.13**	7.89	6.69	42.58	8407.53**
T11	2	66.72	67.72	243.30	134.44	9.55	4.77	41	7906.23**
T12	2	66.28	67.33**	244.76	138.21	4.37	6.23	43.5	8135.13**
T13	2	68.39	69.56	249.70	136.51	6.44	6.46	38.83	8887.99**
T14	2	66.11	67.38**	249.32	138.37	1.90	5.55	41.25	9386.20
T15	2	66.33	68.44	246.42	131.12	6.31	1.75**	41.83	8720.59**
T16	2	67.33	68.72	236.48**	128.27**	3.42	2.21	38.41	8455.61**
T17	2	66.50	67.38**	245.62	133.66	4.90	3.32	44.16	8725.56**
T18	2	67.17	67.89	243.20	135.12	5.38	5.24	41.83	9410.80
T19	3	67.72	69.00	253.07	140.37	9.57	4.58	43.16	8321.90**
T20	3	65.89	67.16**	257.09	143.66	8.22	7.67	40.41	9007.89**
T21	3	67.28	68.00	245.46	138.32	10.41	3.44	41.75	7887.43**
T22	3	67.67	69.33	241.62	130.43**	2.44	6.80	47.5**	7981.56**
T23	3	67.33	68.28	255.39	145.87**	19.08**	5.65	43.41	8857.49**
T24	3	66.33	68.06	258.36	142.52	15.17	2.15**	44	8706.67**
T25	3	67.28	68.61	252.59	142.49	4.78	8.13	45.25	8571.26**
T26	3	65.33**	66.38**	256.87	146.46**	8.90	6.23	47.16**	9068.05**
T27	3	66.94	68.06	248.32	138.60	6.94	5.39	42.58	8648.70**
T28	4	66.94	67.89	258.92**	145.58**	10.42	6.42	41.66	8671.06**
T29	4	66.56	67.44	250.69	134.31	4.33	1.11**	45.5	9075.00**
T30	4	64.11**	64.94**	258.78**	137.09	14.54	6.06	45.75**	9162.40
T31	4	68.83**	69.61	250.68	132.39	18.14**	9.81	41.75	7430.70**
T32	4	66.06	67.50	230.17**	111.07**	1.67	6.67	30.16**	6096.97**
T33	4	66.17	67**	256.41	141.96	10.42	4.21	42.66	9368.61
T34	4	NA	NA	NA	NA	NA	NA	NA	NA
T35	4	65.78	67.33**	248.63	133.36	5.68	0.33**	31.58**	7956.04**
T36	4	67.94	68.83	235.77**	129.17**	6.63	3.14	37.33	7904.00**
T37	5	67.39	68.00	247.57	134.13	7.66	4.08	40.83	9106.41
T38	5	68.17	69.56	243.89	127.67**	6.67	1.50**	35.91	7783.91**
T39	5	65.66**	66.83**	246.99	130.5**	9.34	3.03	38.66	8542.02**
T40	5	65.38**	67.22**	244.46	129.96**	4.24	5.85	39.41	8262.00**

Treatment	*Teosinte source	DTA	DTS	PH	EH	RL	SL	EN	Yield/ha
T41	5	66.72	68.39	242.81	133.03	15.29	3.83	36.83	7791.40**
T42	5	65.5**	66.5**	253.11	135.43	5.64	3.52	42.5	9107.13
T43	5	69.88**	70.33**	255.11	140.19	12.75	8.46	40	8863.92**
T44	5	65.83	66.66**	257.44	143.77	7.32	5.24	45.58**	8801.28**
T45	5	66.56	67.27**	244.83	138.37	9.20	5.44	45.75**	8839.13**
T46	6	66.83	68.17	242.18	130**	8.12	4.07	42.83	8247.39**
T47	6	67.11	68.28	248.80	136.77	6.19	10.73	47.16**	7247.54**
T48	6	66.39	66.83**	244.37	136.56	9.25	1.34**	43.08	8260.44**
T49	6	67.17	68.28	244.07	134.62	4.25	2.41	37.91	6186.95**
T50	6	67.33	68.39	258.18	147.52**	4.34	1.76**	40.58	7709.13**
T51	6	66.67	67.50	245.70	137.88	4.09	6.28	38.66	8216.78**
T52	6	67.83	69.00	250.09	139.89	6.71	0.66**	44	9019.05**
T53	6	67.28	68.56	251.23	142.79	2.33	3.96	44.16	9725.20
T54	6	68.17	68.61	249.29	137.44	5.62	3.08	41.75	8215.20**
T55	7	67.61	68.61	249.42	135.77	8.41	3.63	44.08	9152.62
T56	7	66.72	67.44	251.21	138.53	8.33	10.36	38.25	9294.25
T57	7	64.72**	65.94**	250.12	137.27	3.20	2.17**	42.25	9744.98
T58	7	68.44	69.22	235.2**	128.21**	8.08	4.53	40.91	8988.02**
T59	7	66.28	67.27**	244.58	137.67	8.08	11.31**	44.83	9716.58
T60	7	69.16**	69.17	244.02	137.31	4.73	3.48	39.75	8766.67**
T61	7	68.00	68.61	257.04	142.52	12.43	3.60	42.58	7527.37**
T62	7	68.00	68.72	253.34	139.59	7.96	4.00	40	8430.10**
T63	7	67.00	67.89	257.36	143.53	6.71	2.00**	44.91	9462.74
T64	8	67.11	68.61	247.27	133.92	2.58	4.79	38.25	8028.67**
T65	8	66.83	67.83	251.69	136.81	5.12	3.60	46.25**	8584.03**
T66	8	68.88**	69.56	239.94**	130.98	10.06	3.69	42.5	8958.85**
T67	8	67.78	68.67	245.10	133.83	9.54	6.72	36.91	7503.86**
T68	8	67.17	68.61	237.2**	131.91	6.83	2.48	43.08	8117.22**
T69	8	67.00	68.11	241.15**	134.90	4.51	2.39	44.41	8103.68**
T70	8	66.89	67.89	245.88	137.30	7.68	5.31	43.91	9006.45**
T71	8	68.56	69.22	257.30	143.81	11.83	8.62	43	8655.89**
T72	8	66.00	67.78	247.60	134.41	15.58	5.27	43.25	8783.01**
T73	9	67.61	68.72	242.47	124.54**	4.49	5.01	33.33**	7874.23**
T74	9	66.33	67.83	230.77**	117.93**	8.35	5.39	39.58	7345.99**
T75	9	NA	NA	NA	NA	NA	NA	NA	NA
T76	9	65.78	66.88**	244.34	133.38	3.14	4.35	45.83**	8204.89**
T77	9	68.28	68.89	258.29	143.29	6.54	3.67	45.5	9052.14**
T78	9	66.67	68.17	253.38	141.12	4.78	8.59	43.25	9259.78
T79	9	68.06	69.00	240.23**	132.59	7.38	4.28	38	8354.44**
T80	9	68.83**	69.78	240.87**	131.92	8.99	7.90	49.41**	9120.92
T81	9	66.28	67.83	258.01	138.32	6.87	1.68**	45.16	9212.01

Treatment	*Teosinte source	DTA	DTS	PH	EH	RL	SL	EN	Yield/ha
T82	10	68.06	69.11	255.79	139.96	11.36	6.71	45.83**	9027.39**
T83	10	66.72	67.94	255.14	148.93**	8.85	4.73	49.75**	7119.46**
T84	10	68.17	69.00	261.7**	152.1**	13.11	17.73**	51.58**	9586.70
T85	10	66.56	67.72	251.03	145.62**	6.95	3.42	45.66**	8770.44**
T86	10	68.00	68.56	267.2**	155.41**	10.76	3.87	47.58**	9075.26**
T87	10	67.83	68.83	249.66	146.5**	8.82	8.58	43.33	8372.10**
T88	10	66.72	68.06	267.27**	147.5**	6.82	2**	47.58**	10802.98
T89	10	67.72	68.94	257.18	144.80	7.69	2.20	42.58	10391.32
T90	10	67.33	68.06	262.68**	150.34**	15.58	3.69	48.08**	8522.28**
T91	11	68.83**	69.83	255.83	146.51**	20.30**	4.35	43.08	8606.51**
T92	11	69.44**	70.16**	262.94**	150.73**	16.57**	10.41	46.91**	9530.54
T93	11	69.66**	70.22**	249.11	139.43	4.36	5.16	42.5	8839.82**
T94	11	67.94	69.22	256.94	145.93**	13.84	15.04**	43.33	9042.31**
T95	11	68.39	69.61	252.13	143.76	8.89	4.53	46.66**	8970.66**
T96	11	67.00	68.39	261.86**	146.25**	8.17	2.72	50.25**	9642.95
T97	11	66.78	67.94	263.94**	150.16**	13.83	4.54	43.08	8558.34**
T98	11	70.22**	71.44**	239.46**	131.33	4.50	11.66**	42.33	8013.30**
T99	11	67.44	68.72	267.1**	154.67**	14.28	11.07	53.16**	9165.25
T100	12	66.06	67.33**	266.11**	149.25**	8.75	9.57	43.5	11545.69**
T101	12	68.56	69.67	256.50	144.56	21.78**	6.84	43.33	9542.23
T102	12	69.05**	70.16**	252.41	142.24	18.44**	6.44	41.41	8475.08**
T103	12	67.72	68.94	257.89	153.05**	4.13	4.72	42.75	8579.68**
T104	12	66.83	67.83	255.06	142.33	12.20	0.66**	45.25	8282.74**
T105	12	69.05**	69.72	256.21	147.46**	12.79	9.33	46**	9322.71
T106	12	66.17	68.33	255.10	144.41	10.18	2.67	43.25	8528.67**
T107	12	67.00	69.00	257.57	141.90	17.75**	2.29	47.41**	8984.93**
T108	12	68.83**	69.44	255.96	144.57	6.21	2.92	32**	6925.03**
T109	13	68.28	69.00	257.04	139.43	7.46	3.04	46**	9040.52**
T110	13	67.39	68.00	248.02	130.74	4.22	8.17	39	7749.30**
T111	13	66.06	67.33**	242.50	133.90	4.46	3.74	40.91	7865.64**
T112	13	NA	NA	NA	NA	NA	NA	NA	NA
T113	13	66.50	67.50	247.84	136.58	2.31	5.36	42.75	8364.89**
T114	13	67.78	68.83	245.36	130.71	7.87	5.37	49.16**	8546.96**
T115	13	66.94	67.89	256.89	142.49	8.42	3.87	45.25	9279.73
T116	13	68.56	69.11	235.93**	126.27**	7.27	4.29	35.5	7742.96**
T117	13	NA	NA	NA	NA	NA	NA	NA	NA
T118	14	67.89	69.11	247.34	139.71	6.03	6.22	41.91	7979.62**
T119	14	67.28	68.50	251.88	143.76	13.47	5.82	39.83	7851.15**
T120	14	66.83	67.89	252.06	142.59	5.01	8.65	42.33	7543.48**
T121	14	67.50	68.67	244.27	136.94	5.92	3.64	39.91	8850.45**
T122	14	67.28	67.78	243.78	137.48	5.81	7.02	41.08	8408.68**

Treatment	*Teosinte source	DTA	DTS	PH	EH	RL	SL	EN	Yield/ha
T123	14	67.11	67.83	257.67	141.88	10.66	3.05	46**	9746.98
T124	14	68.61**	69.06	252.62	149.22**	7.53	5.16	48.75**	9208.90
T125	14	65.78	66.44**	256.34	144.73	7.78	13.51**	45.75**	9573.46
T126	14	65.78	66.66**	253.91	149**	18.38**	5.06	42.33	8682.82**
T127	15	66.11	67**	263.1**	144.44	16.23	4.39	43.5	9502.95
T128	15	67.17	68.50	248.70	135.43	11.20	4.47	43.91	8831.64**
T129	15	67.89	68.44	249.73	138.06	6.06	7.83	42.25	8260.59**
T130	15	66.78	67.16**	257.36	143.36	8.33	8.36	43.83	9797.63
T131	15	68.44	68.94	246.82	135.46	5.72	7.50	46.58**	9374.15
T132	15	68.56	69.78	244.76	140.09	5.73	7.20	41.58	9292.45
T133	15	67.67	68.11	268.46**	151.96**	9.38	8.69	42.25	10105.59
T134	15	68.61**	69.89	246.62	135.80	6.39	2.72	42.25	8454.37**
T135	15	65.33**	66.16**	251.48	137.47	6.74	6.82	37.83	9664.10
T136	16	68.77**	69.78	252.14	145.27	16.41**	6.19	46.5**	8619.55**
T137	16	66.22	67.27**	249.21	137.07	7.89	2.51	44.66	9380.95
T138	16	66.94	67.83	241.33**	135.64	4.69	3.49	45.5	8206.61**
T139	16	68.83**	69.50	248.72	137.82	10.14	3.20	44.83	8924.46**
T140	16	67.72	68.28	250.40	139.87	11.72	7.42	44.16	9068.02**
T141	16	66.50	67.78	261.88**	144.20	9.56	3.08	43.08	9192.13
T142	16	66.06	67.16**	249.02	139.17	12.81	4.17	44.66	8750.38**
T143	16	66.17	67.44	251.90	144.56	8.99	3.62	49.58**	9878.51
T144	16	65.27**	65.66**	259.16**	143.11	6.18	1.84**	44.83	9998.67
T145	17	68.00	69.33	245.97	139.23	7.20	4.74	40	7241.08**
T146	17	67.72	68.56	248.74	140.33	7.71	3.36	50.08**	9096.18
T147	17	65.72**	66.72**	257.20	154.55**	9.71	7.24	41.08	8628.62**
T148	17	69.94**	70.22**	247.78	142.26	12.17	1.92**	48**	8692.77**
T149	17	69**	69.39	245.50	136.90	6.77	7.18	37.58	8728.60**
T150	17	65.72**	67.05**	256.34	143.79	12.82	2.26	45	9441.74
T151	17	68.00	69.39	244.04	134.51	4.86	3.82	44.66	8768.01**
T152	17	68.44	69.50	238.68**	134.29	11.28	4.12	36.08	7989.98**
T153	17	67.11	67.89	259.11**	149.06**	11.97	6.55	47.58**	8860.80**
T154	18	68.72**	69.56	257.59	146.8**	5.19	12.24**	44.75	9528.29
T155	18	68.06	68.83	254.62	139.52	8.12	3.37	46.33**	8377.91**
T156	18	69.38**	69.61	243.36	138.56	21.09**	5.07	45.25	8464.80**
T157	18	67.06	68.17	253.57	137.12	9.56	2.52	43.66	9204.13
T158	18	67.39	68.11	250.26	138.37	6.71	2.71	44.33	10534.26
T159	18	66.78	67.50	253.47	143.88	15.26	0.56**	42.66	8518.67**
T160	18	68.11	68.50	259.31**	147.92**	11.15	10.21	49.16**	9328.60
T161	18	68.66**	69.61	258.49	148.25**	16.94**	8.00	42.08	9350.74
T162	18	66.72	67.83	256.76	147.55**	11.22	6.46	41.91	8685.57**
T163	19	66.61	67.89	244.60	136.36	6.49	2.21	38.91	6800.58**

Treatment	*Teosinte source	DTA	DTS	PH	EH	RL	SL	EN	Yield/ha
T164	19	68.61**	69.56	251.10	143.44	13.56	1.70**	50.41**	8167.79**
T165	19	68.28	69.67	253.88	142.57	7.54	3.83	45.91**	8354.04**
T166	19	69.5**	70.27**	237.83**	125.26**	2.03	6.40	37.33	7971.10**
T167	19	68.83**	70.38**	239.15**	128.36**	9.22	2.72	41.5	8651.65**
T168	19	67.00	68.72	250.80	136.13	5.30	1.82**	48.08**	8977.13**
T169	19	68.11	68.89	251.37	134.31	5.58	1.13**	44.08	7731.72**
T170	19	69.83**	70.77**	248.79	136.28	6.31	3.45	41.33	8013.01**
T171	19	68.06	68.33	255.08	143.06	5.21	6.69	46**	8395.53**
T172	20	69.11**	70.00	249.60	140.27	7.33	6.03	40	8133.36**
T173	20	67.39	68.67	267.62**	147.73**	16.76**	2.95	44	8458.41**
T174	20	65.94	67.33**	248.11	134.99	14.05	6.44	35.58	8137.22**
T175	20	68.61**	69.44	243.76	133.42	11.48	5.40	45.83**	8675.23**
T176	20	67.00	68.50	260.23**	148.64**	13.13	4.47	42.16	9344.28
T177	20	67.39	68.89	263.68**	146.93**	10.60	6.42	43.16	9091.51
T178	20	66.83	68.00	261.74**	144.60	14.05	6.41	37.41	9194.24
T179	20	70.27**	71.16**	257.80	152.54**	5.48	7.72	43	7877.88**
T180	20	67.72	68.61	258.57	140.56	5.90	3.23	43.75	9691.92
T185:	Control	67.16	68.73	250.17	137.92	8.17	6.73	39.81	10080.06
T186	CML78xCML311	66.89	68.33	232.01**	129.74**	1.33	3.10	42.58	9473.42
T187	LUG03xCML311	67.56	69.17	229.46**	129.93**	0.67	9.91	42.5	10243.06
T188	DK2027Y	67.44	68.00	249.00	132.54	1.18	2.25	41.41	11620.96**
T189	Cimarron	67.72	67.83	242.07	125.02**	0.33	4.67	41.25	12403.40**
T190	P3055W	64.66**	65.55**	260.06**	136.22	0.52	2.69	43.66	12251.32**
T191	DAS2362	67.39	68.61	249.80	129.48**	1.19	3.69	36.16	10337.21
T192	P3164W	64.72**	65.16**	234.01**	122.37**	0.56	6.08	43.25	12496.33**

** Minimal significant differences to 0.05 probability. *Teosinte source origin can be viewed in table S2. Traits: DTA= Days to Anthesis (d), DTS= Days to Silking (d), PH= Plant Height (cm), EH= Ear Height (cm), RL= Root Lodging percentage (%), SL= Stalk Lodging percentage (%), GW= Grain Weight (kg/plot), Yield/ha= Adjusted Yielding (kg/ha), EN= Ear Number (count) NA= No Available.

Table S2 - Origin of accessions used to form the BC2F1 Maize-Teosinte families which were used as male parents crossed by LUG282 inbred line.

Specie/Landrace	Place of collected population *		Municipality	State/Country
Zea diploperennis +	1 San Andrés Milpillas	(T1-T9)	Huajicori	Nayarit
	2 San Miguel Cuzalapa	(T73-T81)	Cuautitlán de García	Jalisco
Zea luxurians +	3 Las Majadas	(T163-T171)	Jutiapa	Jutiapa, Guatemala
Zea mays ssp. *huhuetenangensis* +	4 El Tablón	(T172-T180)	San Antonio Huista	Huehuetenango, Guatemala
Zea mays ssp. *mexicana* ++	5 Tarahumares	(T10-T18)	Guadalupe y Calvo	Chihuahua
landrace Nobogame				
Zea mays ssp. *mexicana*	6 Potrero El Tepalcate	(T19-T27)	Nombre de Dios	Durango
landrace Durango				
Zea mays ssp. *mexicana* ++	7 El Pedregal	(T28-T36)	Ocoyoacac	State of México
landrace Chalco	8 Opopeo	(T37-T45)	Salvador Escalante	Michoacán
Zea mays ssp. *mexicana* ++	9 El Salteador	(T46-T54)	Yuriria	Guanajuato
landrace Central Plateau	10 Penjamillo de Degollado	(T55-T63)	Penjamillo	Michoacán
Zea mays ssp. *parviglumis* +	11 Camino Carboneras	(T64-T72)	Guachinango	Jalisco
landrace Balsas	12 Crucero Lagunitas	(T82-T90)	Tecoanapa	Guerrero
	13 Los Cimientos	(T91-T99)	Villa Purificación	Jalisco
	14 El Salado-Amates Amarillos	(T100-T108)	Mochitlán	Guerrero
	15 San Cristóbal Honduras	(T109-T117)	San Jerónimo Coatlán	Oaxaca
	16 Huixtitla	(T118-T126)	Amatepec	Guerrero
	17 La Lima	(T127-T135)	Tolimán	Jalisco
	18 Zacatlancillo	(T136-T144)	Teloloapan	Guerrero
	19 El Potrero	(T145-T153)	Huetamo	Michoacán
	20 Plan de los Timbres	(T154-T162)	Huitzuco de los Figueroa	Guerrero

Nine BC_2F_1 families from each initial teosinte accession crossed by CML311 were 450 segregated.
+ teosinte species from southern lowlands
++ teosinte species from northern and central highlands