Genetic Potential and Usefulness of Native Maize Populations in Developing Novel Germplasm for Current and Upcoming Goals

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Abstract

Traditional agricultural system is referring to the maize production based on indigenous or farmers knowledge and practices that have been developed through many generations. In the area of study, genetic maize diversity was explored by the expression of quantitative traits of the ear and the race classification approach. Evaluation results indicated that the native populations adapted to the transition and highland (above 2000 masl) areas, showed a contrasting yield response when they were evaluated at the intermediate environment; whereas, those populations adapted to the lowland and intermediate altitudes showed a satisfactory yield performance in both environments. The above performance pattern is essential because it may be useful to identify favorable alleles that, in a local population *per se* or through genetic combination, results in population changes in allele frequencies that could mitigate the effects of climate changes, particularly in maize populations adapted to highland altitudes. Selection procedures applied to a local adapted population can be managed attending different goals, including the conservation of genetic diversity (*per se* selection), and to develop novel germplasm. The introgression of foreign germplasm into a local population and the application of three selection cycles resulted in a novel variety (JAGUAN) adapted to a regional northeast Mexico environmental conditions.

Keywords: *Zea mays* L., native maize populations, genetic diversity, genetic by environmental interaction, selection procedures

1. Introduction

The maize (*Zea mays* L.) is a native crop of Mexico adapted to the most diverse environmental conditions; planted around the country in altitudes ranging from sea level to altitudes greater than 2550 m. In Mexico, 78.3% of the maize production is sown under rain-fed conditions, mainly with native (landrace) adapted varieties [1]. Those varieties are usually grown in non-optimum agronomic conditions, therefore they are adapted to variable rainfall, and are in some extent, tolerant to biotic and abiotic stresses. These varieties are available to the farmers for sowing, due to their flexible response to adverse situations, and are frequently used for seed exchange among farmers, within the same community or with other communities. In the Coahuila state, 24,900 ha of maize for grain production were sown during 2016, 84.7% was sown in the southeast region, mainly with local adapted populations (landrace populations), and 94.8% of these were sown under rain-fed conditions [1]. Typically, the native maize production is mainly for local consumption, both human and livestock as forage.

The area of study is located in the southeast of the Coahuila state in Mexico, and it is represented by five counties (Arteaga, General Cepeda, Parras, Ramos Arizpe, and Saltillo). Coahuila state is situated in the central part of the North of Mexico, with a territorial area of 151,571 km^2. The climate in the state is dry to very dry, semi warm (75% of total area), average temperature ranging from 18 to 20°C; annual average precipitation of 316 mm. Based on the environmental conditions, the region of study is considered as critical, determined by an average annual precipitation ranging between 350 and 450 mm; average temperature of 16.8°C; with presence of drought and frost seasons in the year. Moreover, to the environmental and ecological conditions in the region, the maize diversity is determined by the adaptation, and genetic combinations among race complexes allowed by seed exchange within and among different communities [2, 3].

Exploring and understanding the genetic potential of adapted cultivars on traits of interest may determine and guide further research for a particular environment or crop system, as well as the efficient use of both economic and human resources. Thus, the objectives of research work were to describe the regional maize genetic diversity, determine the genetic potential of locally adapted maize populations and to identify strategies for crop improvement to resolve current and future aims.

2. Regional maize genetic diversity

In any crop system, the genetic diversity is determined—among other factors—by the use of diverse types of local varieties. Conceptually, two types of genetic materials are commonly developed, those obtained from preferences (color, flavors, crop type, etc.) and those selected for adaptability to biotic or abiotic micro-environments [4]. In addition to the native population for a particular environment, maize genetic diversity is associate to other factors that may change the genetic structure in a population such as the seed exchange among farmers within a community or among different communities, and, depending on the migration index and introgression of foreign germplasm, would contribute to the genetic variation in a native population [5, 6].

The maize (*Z. mays* L.) is a native crop of Mexico, and it is the place where the highest genetic diversity is found. This crop is adapted to the most diverse environmental conditions; thus, the specific local adapted populations (landrace) have been developed, with particular attributes that differentiate each other, within and among regions, circumstances that make possible to recognize Mexico as center of origin and diversification [7]. Commonly, the maize diversity has been described by the racial classification approach, which allowed to identify the first 25 races of maize in Mexico, based on morphological data (plant, ear, and tassel) [8].

In the region of study, the maize diversity has been documented by the presence of representative race populations in Coahuila state, such as Tuxpeño [8], Raton and Tuxpeño Norteño [9], Celaya, Conico Norteño, Elotes Conicos, and Olotillo [10]. A case study carried out in native populations from Coahuila State in Mexico, indicated that genetic diversity shows a continuous pattern among racial complexes, and is associated to the altitudinal and ecological regions [11].

2.1. Relationship among local populations

In this section, several quantitative traits of the ear and grain were considered to analyze the maize genetic diversity and the relationship among local populations within the southeast region of the Coahuila state in Mexico. The racial classifications and relationship among the native adapted populations (landraces) were studied with a sample of 77 maize populations that were collected in the region from altitudes ranged from 774 to 2557 masl. A total of 51 of these populations were collected in 2008 [10] and 26 during 2010 (unpublished data), which represents the maize genetic diversity in the region of study.

Sample sizes of 10 representative ears were first used for a visual classification of the maize populations based on the primary race classification [8]. In addition to the race classification, a set of quantitative traits from the ear and grain were used to analyze the relationship among the native maize populations. Several authors have emphasized that the reproductive organ traits such as the ear traits, are the most useful for race classification in maize [12, 13]. Thus, eight racial complexes were identified: Celaya, Conico Norteño, Elotes Conicos, Elotes Occidentales, Olotillo, Raton, Tuxpeño, and Tuxpeño Norteño. At the same time, maize populations were grouped by an altitudinal stratum: lowland (0–1000 m), intermediate (1001–1800 m), transition (1801–2000 m), and highland (above 2000 m) (**Table 1**).

Ten quantitative ear and grain traits were obtained from the collected sample to analyze the maize diversity. Five ear traits: ear and cob diameter (EAR_DIAM, COB_DIAM) (cm), ear length (EAR_LENG) (cm), ear rows (EAR_ROWS), shelling percent (SHELL_PCT), and five kernel traits: Kernel measurements such as kernel length (KER_LENG), width (KER_WIDTH) and thickness (KER_THICK) (mm), kernel per row (KER_PER_ROW), weight of 100 dry kernel (WT_100_KER) (g) [14]. Data were explored by the analysis of variance to test adaptation groups and racial complexes differences. In both cases, populations within groups and populations within races were analyzed using the PROC GLM procedure of SAS [15]. Data means were used to explore maize diversity by principal component analysis using the quantitative traits as testers [16].

Race classification	**Race ID**	**Lowland (<1000)**	**Intermediate (1001–1800)**	**Transition (1801–2000)**	**Highland (>2000)**	**Total**
Celaya	C		4			4
Cónico Norteño	CN		3	8	23	34
Elotes Cónicos	EC		1	1	1	3
Elotes Occidentales	EO		2			2
Olotillo	O		1			1
Ratón	R	4	16	1		21
Tuxpeño	T		1			1
Tuxpeño Norteño	TN	1	9	1		11
Total		5	37	11	24	77

Table 1. Racial classification of local native populations from the Southeast of Coahuila State in Mexico.

The races Conico Norteño and Elotes Conicos (the ear conical type) are adapted from the transition to the highland areas, in altitudes above 1700 m; whereas, Raton, Tuxpeño, and Tuxpeño Norteño (the ear cylindrical type), the adaptation area is widely: Raton (84–1300 m), Tuxpeño (0–1950 m), and Tuxpeño Norteño (1400–1701) [17]. The maize diversity in the Coahuila state is represented mainly by three racial complexes: Conico Norteño, Raton, and Tuxpeño Norteño [10].

The eight racial complexes and the four adaptation groups were statistically different ($P \leq 0.01$) for most traits, indicating relative differences among the race type and the adaptation of landrace populations in the region; the populations within racial complexes and populations within adaptation groups were significant ($P \leq 0.01$), indicating the variation associated within race groups, the genetic combination among populations and race complexes (**Table 1**), and the specific adaptation to the different ecological environments within the region.

The scatter plot of the interaction among the 77 native maize populations with the 10 quantitative traits is presented in **Figure 1**.

The maize populations and the traits studied are all distributed along **Figure 1**, where individual points (maize population or traits) reached a vector from the origin indicates the joint association that makes the distinctions among maize populations and the relationship with the associated traits. The group of populations indicated by the dashed oval corresponds to the Conico Norteño and Elotes Conicos, two races adapted to highland altitude, characterized by a conical ear type. Populations outside the oval show the cylindrical ear type represented mainly by the races Raton, Tuxpeño, and Tuxpeño Norteño (**Table 1**). By the exploration of dispersion of these population × racial groups in **Figure 1**, it is possible to detect a continuous pattern among the races Raton and Tuxpeño Norteño. Similar genetic variation pattern was found among racial groups through the landraces analyses in the state of Coahuila [11].

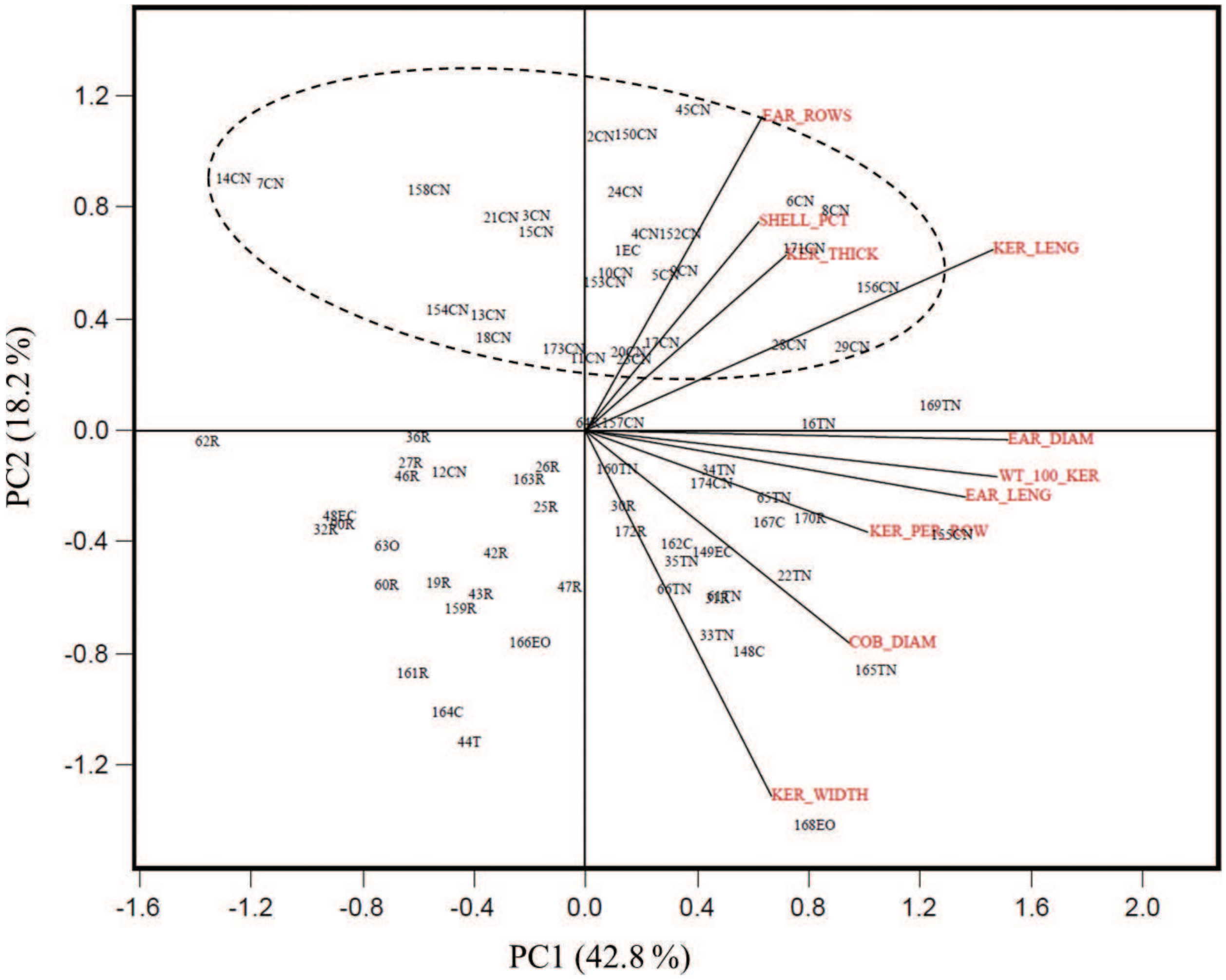

Figure 1. Scatter plot of the first two principal component scores showing the dispersion of the 77 maize populations and relationship among traits. Populations are identified by the population number and combinations of letters that indicate the race classification: C = Celaya; CN = Conico Norteño; EC = Elotes Conicos; EC = Elotes Occidentales; O = Olotillo; R = Raton; T = Tuxpeño; and TN = Tuxpeño Norteño.

Dispersion in **Figure 1** shows the identification of two main groups: the group of maize populations with the conical ear type (transition and highland altitudes) and those with the cylindrical ear type (lowland to intermediate altitudes) (**Table 1**). This indicates the usefulness of the ear and grain traits in exploring the association among maize populations as suggested by [12, 13]. The continuous pattern among the different races within the two main groups is explained by the adaptation of the local populations (**Table 1**), and the genetic combination among races as a consequence of seed exchange by farmers within the community and with other communities [5, 6]. The seed exchange among farmers is a common practice in the area of study, is evident by the presence of some maize populations located at a different adaptation area (**Table 1**), explained by the genetic combinations among different races, and further adaptation to micro-environments as was verified in several regional studies [3, 11].

3. Genetic potential of maize populations

Maize genetic diversity accounted by the locally adapted populations (landraces) within a traditional agricultural system is not static; it varies from 1 year to the another as a consequence of many factors such as migration (seed, pollen), selection, genetic drift [18], and adaptation to changes and interactions with external factors within the ecosystem development, as part of the evolutionary process and selection [19]. Thus, genetic variation is closely related to the environmental and production conditions and to the different uses of the crop, in particular the grain (color and flavor). The knowledge and understanding of the genetic variation, the environmental interaction and its potential use, may determine both the genetic conservation strategy and the possible utilization in breeding programs to improve local populations or for developing novel germplasm for particular goals.

3.1. Yielding potential and environmental response

The maize genetic diversity in the Coahuila state was determined initially by the description of 90 native maize populations that were collected during 2008 [10]. At the same time, those populations were established on field experiments for agronomic evaluation for 2 years (2008–2009), at two contrasted locations to determine the grain yielding potential. The agronomic evaluation was conducted in: El Mezquite, Galeana, Nuevo Leon (1890 masl), and General Cepeda, Coahuila (1350 masl). These locations are representative of both, the highland and intermediate environmental conditions in the area of study. The combination of two locations and 2 years of evaluation was named as four different environments. To analyze the environment response, the native populations were grouped based on the adaptation altitude: lowland (0–1000), intermediate (1001–1800), transition (1801–2000), and highland (greater than 2000 masl).

The local population × environment interaction analysis allowed to identify three groups that describe the specific adaptation of the maize populations [20]: the first one with adaptation to El Mezquite (33.3%), the second adapted to General Cepeda (42.2%), and a third group (24.4%) with an average yielding potential across environments, indicating a form of stability [16, 21]. In this study, it was shown that the racial types are associated to the locations of evaluation: the race Conico Norteño to El Mezquite (highland) and the races Raton, Tuxpeño, and Tuxpeño Norteño to the General Cepeda site (intermediate), which are also associated to the adaptation origin as indicated in **Table 1**. In Mexico, the maize genetic diversity is associated to the agro-ecological conditions that determine the different races types and their ear and grain distinctiveness and uses [17]. Results of the study determined the yielding potential and population response to the environmental evaluation, outstanding the racial groups Tuxpeño, Tuxpeño Norteño, and Raton with the highest yielding potential.

The assigned groups of native maize populations and the groups × environments were statistically different ($P \leq 0.01$), explained by the diversity in altitudinal origin of maize populations and the differential response on the evaluation environments. A relative comparison of the average grain yield of the groups of native populations evaluated in the two contrasted environments during 2 years is presented in **Figure 2**.

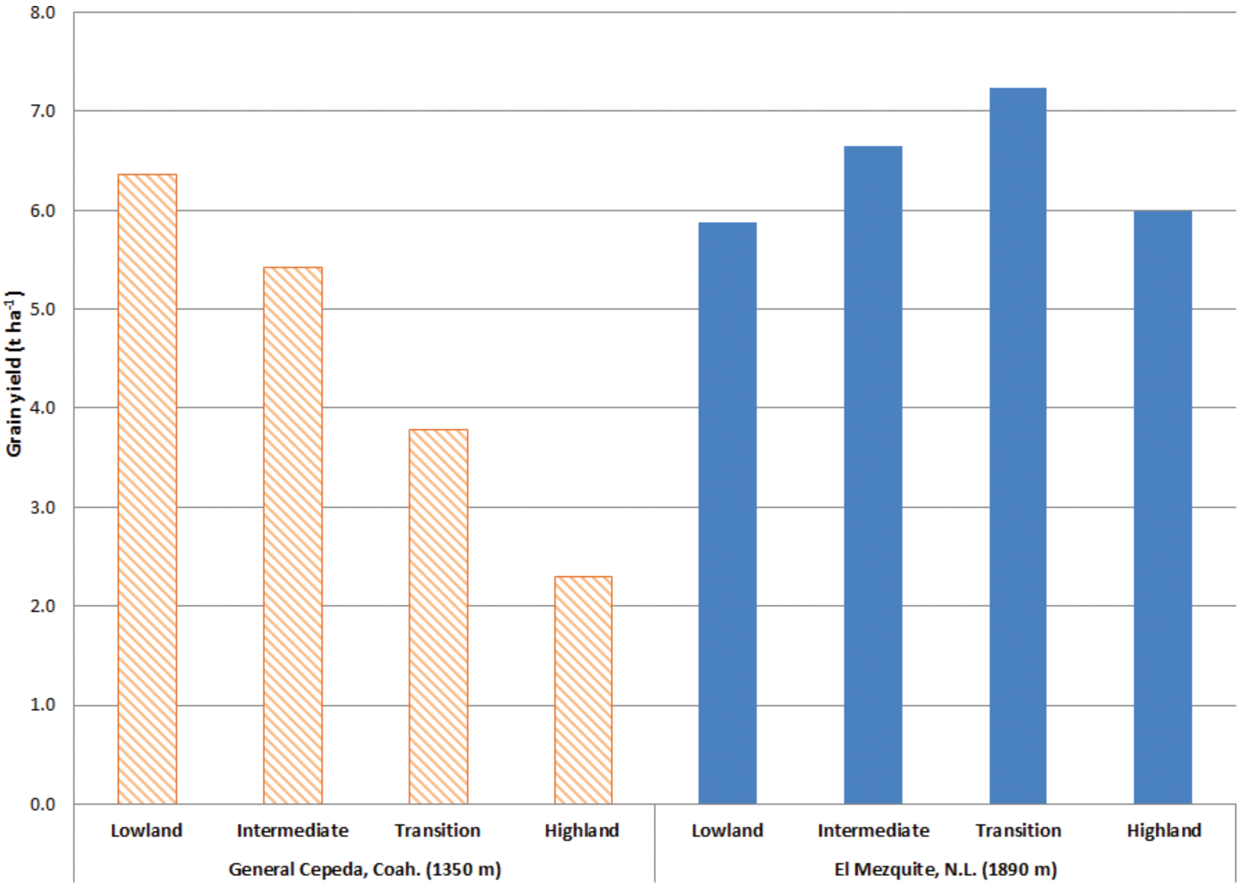

Figure 2. Groups of native maize populations by the environment interaction based on grain yield.

The maize populations represented in the transition and highland groups, showed a contrasted yielding response when evaluated at the General Cepeda area (intermediate altitude), in comparison with El Mezquite environment; whereas those populations adapted to the lowland and intermediate altitudes showed an adequate yield performance in both environments. Similar response pattern has been reported by [22], who also mentioned that populations adapted to highland areas have a difficult performance when established at lowland altitudes; on the other hand, those populations with an adaptation to lowland to intermediate areas have an acceptable agronomic performance. Thus, populations from lowland and intermediate altitudes have better adaptation range with satisfactory yielding potential. The above performance pattern is important because it may be useful to identify favorable alleles that, in a local population *per se* or through genetic combination among different racial complexes, resulting in population changes in allele frequencies controlling traits of interest, consequently, it could mitigate the effects of climate changes, particularly in maize populations adapted to highland altitudes [23].

In a different study, carried out in the southeast of the Coahuila state, an agronomic evaluation of native maize populations was performed in 2013 (unpublished data). The objectives of the research work were to determine the agronomic performance and yield potential of local maize populations, and to define the area of adaptation using two contrasting and representative environments of the southeast of the Coahuila state in Mexico. The agronomic evaluation of 63 maize populations and 7 improved checks was carried at 2 locations and 2 replications (blocks) under irrigation conditions: El Mezquite, Galeana, N. L. (1890 masl) and General Cepeda, Coah. (1350 masl). The combination of two locations and two replications was named as four different environments (GC1, GC2, MEZ1, and MEZ2). In both locations, replications were established independently, and in General Cepeda, the two replications represented two planting dates. The genetic diversity was represented by eight racial complexes: Celaya (3), Conico Norteño (26), Elotes Conicos (4), Elotes Occidentales (1), Olotillo (3), Raton (16), Tuxpeño (6), and Tuxpeño Norteño (4). The improved materials used as checks have variability on maturity and grain type: an experimental variety (POBAM), two improved varieties (VAN210 and JAGUAN), and four synthetic populations (6221, 6222, Pool31, and Pool32). The yield potential was analyzed across environments and the genotype × environment interaction based in the model of the additive main effects and multiplicative interaction (AMMI) [24].

The analysis of variance showed differences ($P \leq 0.01$) among environments, genotypes (populations and checks) and genotype × environment interaction. Among the 25 outstanding populations, the racial groups with higher yield grain potential correspond mainly to the Raton races (9 populations), Tuxpeño (6 populations), and Tuxpeño Norteño (4 populations). Also, there were five native maize populations adapted to intermediate areas: three of the Tuxpeño race (I38T, I52T, and I54T) and two of the Raton race (I13R and I40R) with similar yields to the best improved check. In a previous study, the races Raton, Tuxpeño, and Tuxpeño Norteño had also the highest yield potential [20].

In the environmental response analysis based on the AMMI model, (**Figure 3**) shows the main effects for grain yield (Genotype and Environment) on the abscissa axis, and the first interaction principal component (IPC1) on the ordinate axis.

The AMMI model (**Figure 3**) allowed identifying genotypes with specific adaptation to the two contrasting environments, and the average response across environments. For instance, using an approximate range of $-0.25 \leq 0 \leq 0.25$ of the IPC1 as criteria to identify those genotypes with an average performance across environments, represents a form of stability stability [21]. There were five populations identified with good yield potential and a type of stability across environments: two Conico Norteño (H28CN and H27CN), one of Tuxpeño Norteño (I59TN), and two of Raton race (I56R and I40R). Likewise, in addition to the two Conico Norteño populations (H43CN and T08CN), there were four populations with adaptation to intermediate areas, with good potential in high altitude valleys: Tuxpeño race (I52T and I54T), a population of Elotes Occidentales (I33EO), and another Raton population (I35R). Most of the populations with the highest grain yield on **Figure 3** (positive values of main effects) have adaptation to intermediate areas, and have a potential performance in the two contrasting environments and the stability as well. This pattern agrees with the results presented in **Figure 2**, suggesting that those native populations may be used in a breeding strategy, individually, or as a combination with populations adapted to highland altitudes to identify useful alleles for developing novel germplasm that could mitigate the climate change.

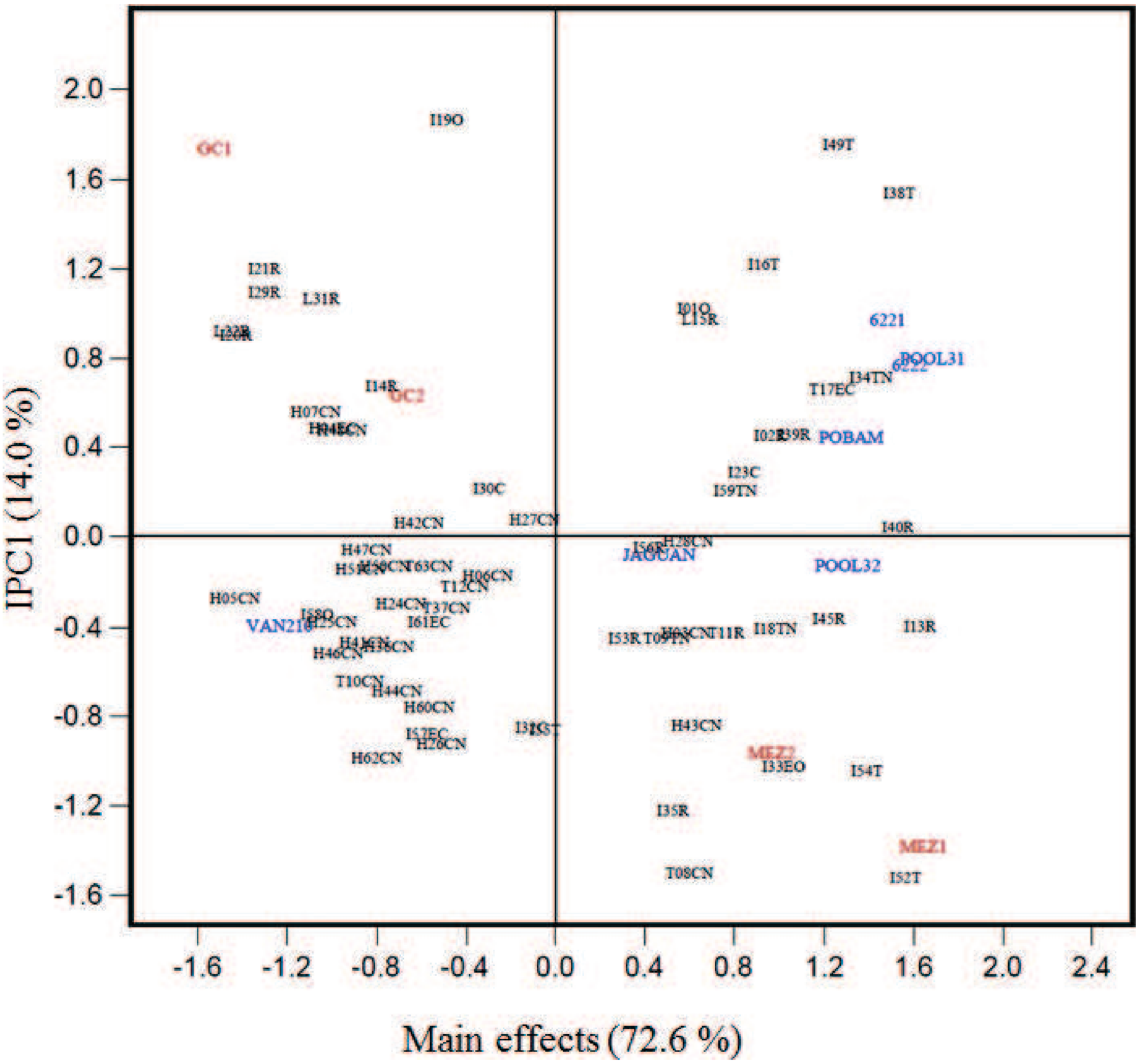

Figure 3. Scatter plot of AMMI model for grain yield of 70 maize genotypes evaluated at 2 contrasting environments (GC1, GC2, MEZ1, and MEZ2). Population's data points are indicated by combinations of letters and numbers. The first character indicates the adaptation area L = lowland, I = intermediate, T = transition and H = highland; followed by the population number and finally, the racial group: C = Celaya, CN = Conico Norteño, EC = Elotes Conicos, EO = Elotes Occidentales, O = Olotillo, R = Raton, T = Tuxpeño, and TN = Tuxpeño Norteño.

4. Strategies for crop improvement

In a traditional agricultural system, the native maize populations are developed and maintained by farmers through multiple cycles of empirical mass selection. Those populations are commonly the only source of genetic variation available for sowing, due to their flexible response to adverse situations, and usually two types of local varieties may be distinguished: the local varieties that are planted in a very small area for special uses, basically for consumption, and represent the diversity of the crop; whereas, in the second case, varieties are planted in a larger areas, widely distributed, and are frequently used for seed exchange among farmers within the same community or with other communities. In either case, strategies for the efficient use of the germplasm need to be determined. For instance, in the first case, the recurrent selection strategy applied is basically to improve it; while, the second group, in addition to *per se* selection, those varieties are eligible for any form of genetic combination with external exotic germplasm.

4.1. Selection strategies applied to a locally adapted maize population

In the southeast of the Coahuila state, a wide adapted local maize population was identified to apply different selection strategies [25]. The native variety named JAGUEY, representative of the race Conico Norteño, is adapted to Jagüey de Ferniza, Saltillo, Coahuila (2100 masl). This variety was exposed to different management and selection procedures where four populations were obtained: (1) the original adapted population (OP); (2) the first generation from the local population (G1), obtained through a seed production scheme (detasseled rows); (3) and (4) two populations generated by the combination of the original population with an improved population, using a divergent selection for early (EM) and late (LM) maturities, respectively. After the populations were developed, a set of 25 half sib families was randomly obtained from each of the 4 populations for evaluation in 2 locations during 2003: El Mezquite, Galeana, Nuevo Leon (1890 m) and Jagüey de Ferniza, Saltillo, Coahuila (2100 m), being the irrigated and rain-fed environments, respectively. The four populations were compared to analyze the effects of selection procedures on agronomic traits using the site and the local population as references. Data were recorded for days to anthesis, plant height (m), husk cover (%), stalk and root lodging (%), moisture content of seed (%), number of ears per plant, and ear yield (t ha^{-1}) adjusted to a 15% moisture content.

Results showed significant differences ($P \leq 0.01$) among populations for most traits. A relative comparison among pairs of populations from a multivariate analysis, based on the agronomic traits evaluated is presented in **Table 2**.

A relative comparison among pairs of populations based on a multivariate analysis, showed significant differences ($P \leq 0.01$) among the OP and G1, with the two populations obtained by the introgression with improved germplasm, in the two environments evaluated (EM and LM), indicating the contribution of the improved material to the original population. On the other hand, there was not any evidence of a difference among the OP and the G1; at the same time, they were comparatively more diverse than EM and LM, as an effect of the selection methodology. Thus, populations showed significant differences in the agronomic traits, determined by both, procedures and selection applied criteria, which determine the selection strategy and management for the conservation and use of genetic diversity.

Contribution of selection methodologies after the first selection cycle, indicated by the average difference in grain yield between G1 and OP, was 1.7%; whereas, the contribution associated to the germplasm combination, the EM against OP was in the order of 24.0%. In both cases, the first cycle of selection was associated with reduction in root and stalk lodging percentages, asynchrony silk interval, husk cover, and plant and ear height, in reference to the original

Populations[†]	G1	EM	LM
	Jagüey, Saltillo, Coahuila (Rain-fed)		
OP	0.338	3.636**	11.396**
G1		2.850**	12.267**
EM			9.844**
	El Mezquite, Galeana, Nuevo Leon (Irrigation)		
OP	0.609	3.219**	11.258**
G1		2.806**	10.628**
EM			6.462**

[†]**, Significant at 0.01 probability level; OP = Original local population; G1 = Fist generation obtained through a seed production scheme; EM and LM = Early and Late maturity populations obtained through the local × improved germplasm; adapted from Rincón and Ruiz [25].

Table 2. Squared distances among pairs of maize populations based on agronomic traits evaluated at two environments.

population. These results indicate that genetic variation of local populations can be managed attending different goals including conservation of genetic variation and crop improvement by the introgression of exotic germplasm.

4.2. Potential of local × improved combination to enhance a locally adapted maize population

Based on the results of the research paper carried out by [25], plants of the local population JAGUEY were crossed with an improved population to determine the value of a breeding material to enhance a local adapted maize population. Introgression of exotic germplasm to adapted material in maize has been a powerful tool to increase genetic variability in the local population, as well as to transfer favorable alleles, such as insect or disease resistance. The proportion of the exotic germplasm has been addressed in several studies to determine the usefulness of the foreign material on the foundation of breeding base populations [26–28]. The relevance of the introgression of foreign or exotic germplasm to an adapted population may change depending on the particular objectives. For instance, the improvement of a local farmer population by the introgression of exotic or foreign germplasm requires the identification of a good source donor and the establishment of the selection strategy. However, the application of breeding techniques to local maize populations may change their genetic structure, being the level of change related to the breeding methodology and the selection pressure. Besides the improvement of the local material, it is essential to preserve as much as possible the genetic variation accounted by the local material. In this case, the maize population JAGUEY adapted to Jagüey de Ferniza, Saltillo, Coah., Mexico, was considered the local material (L), and used to assess the contribution of an improved material introgressed to the local population [25]. This local population is a white dent type of the race Conico Norteño, adapted to rainfall conditions, such as low fertilizer inputs and limited water supply, and it is maintained by farmers through an empirical mass selection. The improved material (I), considered the foreign material, was an early flowering experimental population (CPRE), previously chosen, based on its combining ability performance when crossed with local populations [29]. Initially, full sib (FS) families were obtained throughout plant-to-plant crosses between plants from the local (JAGUEY) and the improved (CPRE) maize populations [29]. Derived families evaluation data included days to flowering (anthesis and silking), plant and ear heights (m), husk cover (%), stalk and root lodging (%), number of ear per plant, and grain yield (t ha^{-1}) adjusted to a 15% moisture content. Best families (10%) were selected based on the evaluation data under irrigated and the rain fall conditions, with special emphasis on the performance under the rainfall environment [30]. After the first genetic recombination of selected families (50:50 of local and improved germplasm), three full sib selection cycles were applied to develop an improved native maize variety named JAGUAN [31]. In addition to grain yield, the selection procedure included number of ears per plant, husk cover, with special attention in keeping the same flowering date as the original native population using selection indices [32]. The difference in grain yield between JAGUAN and the local variety JAGUEY, the original population was 24.5%. JAGUAN is a variety developed for rain-fed conditions, with intermediate biological cycle (83–90 days flowering), plant height of 2.5 m, selected by high planting densities (50,000–60,000 plants ha^{-1}), adapted to the transition to highland areas (above 1800 m), and with phenotypic expression as the original native population [29].

5. Conclusions

Regional genetic maize diversity was explored based on the race classification approach using selected ear and grain traits. Eight race complexes were identified that represent the maize diversity in the region of study. There are local populations with a wide and specific adaptation; for those wide adapted populations a crop improvement selection scheme was applied, whereas, on the specific adapted populations, a strategy for conservation and use could be implemented. Genetic introgression to a native population and further selection criteria has been useful to develop a novel variety JAGUAN adapted to regional environmental conditions. Genetic combinations among selected populations that represent the genetic diversity within the region have been identified as potential allele donors to improve genetic materials to mitigate the alterations associated to the climate change effect, particularly in populations adapted to highland altitudes.

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