

Maize landrace diversity persists in rural areas in transition to urban areas

La diversidad de maíces nativos persiste en áreas rurales en transición hacia lo urbano

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ABSTRACT

Among the factors that are known to threaten the diversity of maize landraces is increasing urbanization, as it causes the reduction of agricultural areas and contributes to the loss of varieties. Although the urbanization process is occurring in several rural areas, there are currently few studies aimed at determining the level of morphological diversity present in the maize landraces still cultivated in these areas and at determining the association degree with the races reported therein. This was the objective of this research. Therefore, during the year 2021, in the Coronango municipality, Puebla, 54 maize landraces were collected which, along with three racial controls, six experimental varieties, and a commercial control, were evaluated under rainfed conditions in three auxiliary boards of the municipality, using a simple 8×8 lattice design. Thirty-seven variables (phenological, morphological, and derived indices) were quantified and subjected to a combined analysis of variance, cluster and principal components analyses. There were highly significant differences among materials for the 37 evaluated variables. Multivariate analysis revealed the existence of three groups of landraces, differentiated from each other basically by grain color, earliness, and characteristics of leaf, tassel, ear, and kernel. These groups showed little relationship with the Cónico and Elotes cónicos races, with the exception of one of the white grain groups, that was associated with the Chalqueño race. It is concluded that, despite the pressures faced by this area in transition between rural and urban conditions, an important level of morphological diversity persists among the maize landraces cultivated therein.

KEY WORDS: Peri-urban agriculture, genetic diversity, maize landraces, urbanization.

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RESUMEN

Entre los factores que se afirma amenazan la diversidad de maíces nativos está la creciente urbanización, pues ocasiona la reducción de áreas agrícolas y contribuye a la pérdida de variedades. Aun cuando el proceso de urbanización está ocurriendo en varias áreas rurales, actualmente son escasos los estudios orientados a precisar el nivel de diversidad morfológica presente en los maíces aún cultivados en tales espacios y a determinar su grado de asociación con las razas ahí reportadas. Ello constituyó el objetivo de esta investigación. Por tanto, durante el año 2021, en el municipio de Coronango, Puebla, se recolectaron 54 poblaciones nativas que, junto con tres testigos raciales, seis variedades experimentales y un testigo comercial, se evaluaron bajo temporal en tres juntas auxiliares del municipio, utilizando un diseño látice simple 8×8. Se cuantificaron 37 variables (fenológicas, morfológicas, e índices derivados), las cuales se sometieron a análisis de varianza combinado, de conglomerados y de componentes principales. Hubo diferencias altamente significativas entre materiales para las 37 variables evaluadas. El análisis multivariado reveló la existencia de tres grupos de poblaciones nativas, diferenciados entre sí básicamente por coloración del grano, precocidad y características de hoja, espiga, mazorca y grano. Tales grupos mostraron escasa relación con las razas Cónico y Elotes cónicos, pero uno de los de grano blanco se asoció con la raza Chalqueño. Se concluye que, pese a las presiones que enfrenta este espacio en transición entre lo rural y lo urbano, aún persiste un nivel importante de diversidad morfológica entre los maíces ahí cultivados.

PALABRAS CLAVE: Agricultura periurbana, diversidad genética, poblaciones nativas de maíz, urbanización.

Introduction

Mexico is worldwide recognized as the geographic region where maize originated and domesticated, as well as an important center of diversification (Ortega *et al.*, 2013). Evidence of the latter is the existence of 68 races that have been reported so far, distributed throughout the country (Caballero-García *et al.*, 2019). According to Ortega (2003), each of these races is formed by multiple landraces, maintained by farmers who sow and select them after each agricultural cycle. For this reason, it has been pointed out that peasants and indigenous people are the “guardians” of the wide diversity of native maize and that they play a fundamental role in its conservation and diversification (Serratos, 2009).

Maize diversity is threatened by multiple factors that contribute to genetic erosion (which includes, among other aspects, the loss of varieties): agricultural modernization, maize imports, the use of flour instead of grain, migration, natural disasters, social conflicts (Orozco-Ramírez &

Astier, 2017), and the use of improved varieties (Van de Wouw *et al.*, 2010). Yet another factor is represented by increasing urbanization, as it implies population displacement and occupation of agroecological zones, leading to the progressive reduction of agricultural land (Martínez & Monroy-Ortiz, 2009) and resulting in the abandonment of maize landraces in many areas (Guzzon *et al.*, 2021).

In Mexico, Puebla is one of the states that significantly contributes to national corn production. It ranks eleventh in production and in 2021 had a harvested area of 497,476.80 ha (97 % of the total sown in the state) with a production of 1,151,896.04 t and an average yield of 2.32 t·ha⁻¹ (SIAP, 2021). However, according to Martínez & Monroy-Ortiz (2009), Puebla state is one of the four entities where urban growth for the period 1995-2005 reached 1,200 km² (the average for the rest of the country was 200 km²). Such growth occurred mostly in detriment of productive zones, adjacent to urban centers, particularly those of rain-fed agriculture. These trends may continue today, so there is likely an impact on the diversity of maize cultivated in agricultural areas near the cities.

In the state of Puebla, studies that have been conducted to determine the levels of morphological diversity and racial types in maize landraces have focused on important or little-explored micro-regions in terms of maize production, such as the Puebla Valley (Hortelano *et al.*, 2008), the Central-Eastern Highlands (Hortelano *et al.*, 2012), the Northeastern Sierra (Contreras-Molina *et al.*, 2016), the Humid Tropics (López-Morales *et al.*, 2014), the Western Highlands (Alvarado-Beltrán *et al.*, 2019) or the entire Highlands (Flores-Pérez *et al.*, 2015). Consequently, at the moment, there is no research directed to analyze the level of diversity of maize landraces still cultivated in areas that are in an urbanization process. Therefore, the objective of this study was to analyze the level of morphological diversity currently existing among maize landraces cultivated in a rural area undergoing urbanization, and their relationship with the races reported for that area. The study provides information on the level of morphological diversity present in an area with factors that put it at risk.

Materials and Methods

Study area

The study was conducted in the Coronango municipality (19° 05' 53.88" to 19° 10' 26.76" N, 98°15' 32.04" to 98° 19' 24.60" W and 2,190 masl), located in the Puebla Valley (INEGI, 2020). The municipality is made up of three auxiliary boards (San Antonio Mihuacán, San Francisco Ocotlán, and San Martín Zoquiapan) and the municipal seat (Santa María Coronango). According to INEGI (2010) and INEGI (2023), the climate is temperate sub-humid with summer rains; the average annual temperature is 14-18 °C and rainfall ranges between 800 and 1000 mm; two types of soil (Arenosol and Phaeozem) predominate in the area. Currently, urban areas are growing on land previously dedicated to agriculture.

Collection and genetic material

The collection of maize landraces was carried out between February and April 2021 in the Coronango municipality. The sampling frame was constituted by the list of peasants in the Coronango municipality registered as corn producers in the Producción para el Bienestar Program spring-summer 2020 cycle, made up of 207 peasants. With the information from this frame, a stratified random sampling technique with Neyman distribution (Singh & Mangat, 1996) was applied, considering boards and head of the municipality as strata. The accuracy was set at 10 % concerning the mean, and the confidence ($z_{\alpha/2}$) at 95 %. The sample size obtained was 63 peasants, distributed as follows: Coronango: 4, Mihuacán: 13, Zoquiapan: 14, and Ocotlán: 32. In each site, from the municipality register of peasants, ordered by auxiliary boards and municipal seat, the number of farmers corresponding to the one determined for each stratum was randomly chosen; once the peasant's names were known, they were contacted to explain them the purpose of the study and maize samples were collected with those who agreed to provide them.

As a result of the collection, 54 samples of maize landraces were obtained (Table 1), which are grown under strictly rainfed conditions, with sowing dates concentrated in the month of May. All of them were included in the evaluation. Three accessions from the CIMMYT germplasm bank, representative of the Chalqueño (CIMMYTMA1398), Cónico (CIMMYTMA10232), and Elotes cónicos (CIMMYTMA1895) races, were used as racial controls. Additionally, six experimental materials of different grain color and earliness were included (three synthetics of white grain: 'Nopalucan Precoz', 'Nopalucan Ultraprecoz' and 'Serdán' and three composite varieties: LHM Blanco, LHM Amarillo, and LHM Azul) and one commercial variety (Niebla®).

Table 1. Maize landraces collected and evaluated in the municipality of Coronango, Puebla, 2021.

Collection site	Grain color					Altitude (m)
	White	Blue	Yellow	Pinto	Red	
Santa María Coronango	4 [†]	2	2	-	-	2187
San Antonio Mihuacán	5	2	-	1	-	2198
San Martín Zoquiapan	8 ^{††}	4	2	-	-	2189
San Francisco Ocotlán	14	6	1	1	2	2190

[†]Includes a sample from a creolized variety; it was identified as C003(WN); ^{††}Includes a sample of cacahuacintle-like maize; it was identified as Z027(C).

Experimental evaluation

To evaluate the 64 materials, a simple 8×8 lattice was used. The experimental unit consisted of two rows 5 m long and 0.85 m wide, with eleven hills per row, spaced every 50 cm. The useful plot was the total of the experimental unit. Experiments were established in three locations in the Coronango municipality: San Francisco Ocotlán (19° 08' 43.0" N and 98° 16' 20.0" W), San Martín Zoquiapan (19° 08' 10.0" N and 98° 18' 17.0" W) and San Antonio Mihucán (19° 08' 34.0" N and 98° 18' 29.0" W) (INEGI, 2020); sowings were carried out on May 19, 21 and 24, 2021, respectively.

Experiment management

At the time of sowing, three seeds were deposited per stroke, and 35 days later, thinning was made by two plants per hill. Sowing and crop management were carried out according to the conventional practices of farmers in the region, except for fertilization, for which a dose of 110N-60P-00K was used, applied fractionally: 40N-60P-00K in the first tillage (27 days after sowing) and the remaining nitrogen in the second tillage (40 days after sowing), using diammonium phosphate and urea as sources. Weed control was carried out with tillage and, after the second tillage, with the application of 2,4-D amine and atrazine at a rate of 1 L·ha⁻¹.

Variables recorded

The variables days to 50 % tasseling (DTT) and silking (DTS) were quantified considering all the experimental unit, as described by Rocandio-Rodríguez *et al.* (2014). Once flowering was concluded, five plants with complete competition were randomly chosen in each plot, in which the vegetative variables described in Table 2 were recorded, as described by Hortelano *et al.* (2012). The tassels of the five plants mentioned were collected to measure the listed attributes, following Rocandio-Rodríguez *et al.* (2014). Finally, at harvest, a sample of five representative ears was taken from each plot, to measure in them and their grains the corresponding characteristics, as explained in the methodology described by Hortelano *et al.* (2012). In addition, several indices were calculated, derived from the variables contained in Table 2.

Table 2. List of measured variables and calculated indices in the studied maize populations. Coronango, Puebla, 2021.

Type of variable	Variable	Abbreviation	Unit of measure
Phenological	Days to tasseling	DTT	days
	Days to silking	DTS	days
Vegetative	Plant height	PLANTH	cm
	Ear height	EARHT	cm
	Total leaves	TOTLEAV	number
	Leaves above the uppermost ear	LEAVA	number
	Leaves below the uppermost ear	LEAVB	number
	Ear leaf length	LEAFLT	cm
	Ear leaf width	LEAFWD	cm
	Leaf area [†]	LAREA	cm ²
	(EARHT / PLANTH) Index	IHEIGHT	dimensionless
	(LEAVA / LEAVB) Index	ILEAVES	dimensionless
Tassel	Peduncle length	PEDLT	cm
	Length of the tassel branching space	LTBRS	cm
	Length of the central branch	LCENB	cm
	Total tassel length	TTSLT	cm
	Length of the basal lateral segment	LBLS	cm
	Primary branches	PRIMB	number
	Secondary branches	SECB	number
	(LTBRS / TTSL) Index	ILTBTS	dimensionless
	(LCENB / TTSL) Index	ILCBTS	dimensionless
	(PEDL / TTSL) Index	ILPEDTS	dimensionless
(TTSL / PLANTH) Index	ITSLPHT	dimensionless	
Ear	Ear length	EARLT	cm
	Ear diameter	EARDM	mm
	Row number	ROWNO	number
	Kernels per row	KRROW	number
	(EARLT / EARDM) Index	IEAR	dimensionless
Grain	Kernel length	KRLT	mm
	Kernel width	KRWD	mm
	Kernel thickness	KRTK	mm
	Kernel volume ^{††}	KRVOL	mm ³
	Hectolitre weight	HECWT	kg hL ⁻¹
	(KRLT / KRWD) Index	IGRAIN	dimensionless
	(KRWD / KRLT) Index	IWDLT	dimensionless
	(KRTK / KRLT) Index	ITKLT	dimensionless
(KRTK / KRWD) Index	ITKWD	dimensionless	

[†]Leaf area = LEAFLT×LEAFWD×0.75; ^{††}Kernel volume= KRLT×KRWD×KRTK.

Statistical analysis

A combined analysis of variance was performed for the 37 variables recorded and then, a Pearson correlation analysis was applied to identify pairs of correlated variables ($r \geq |0.7|$), from which, for the subsequent multivariate analysis, the one considered most informative was chosen. With the averages per population for the 27 resulting variables, a cluster analysis was performed, based on a Gower distance matrix, using Ward's Minimum Variance as a grouping method. In the resulting dendrogram, using the pseudo-F criterion, four groups were identified, for which a multivariate analysis of variance was performed, followed by an analysis of variance by groups (considering the materials included as replicates) and a Tukey mean comparison test ($P \leq 0.05$). Finally, a principal component analysis was also performed, from which a distribution graph of the materials was generated based on the values of the first three principal components, including the groups identified in the dendrogram. All statistical analyses were performed with the SAS OnDemand for Academics® program (SAS Institute Inc., 2012-2020).

Results

The collection consisted of 54 landraces, 31 of white grains (including one of cacahuacintle type maize), 14 of blue, five of yellow, two of red, and two of pinto grains. Of the localities of collection, it was in Ocotlán where samples of all coloration groups were found.

The analysis of variance (Table 3) revealed that among environments there were statistical differences in 32 of the 37 variables studied, indicating that the level of expression in most of the variables depended on the evaluation site. Zoquiapan was the site where the highest statistical values were obtained for attributes such as ear and plant height (258.1 and 149.8 cm, respectively), ear leaf width (9.3 cm), number of secondary branches on the tassel (1.46), as well as ear length and ear diameter (15.1 cm and 48.3 mm, respectively), number of rows and kernels per row (15.4 and 31.2, respectively) and kernel length and width (14.9 and 8.0 mm, respectively). It was followed by Mihuacán (with plant height, ear leaf width, ear diameter, and number of rows of the ear statistically equal to those of Zoquiapan), and finally Ocotlán.

Highly significant differences were observed among materials in all measured variables, suggesting that, among landraces and controls, there are still important diversity levels in all the characters and indices studied. The interaction materials×environments was not significant for 97 % of the variables, which indicates that the behavior of the studied maize populations, in terms of the expression level of most of its attributes, remained stable across environments.

Table 3. Mean squares of the combined analysis of variance of 37 variables of maize landraces, racial controls, experimental varieties and one commercial variety evaluated in the municipality of Coronango, Puebla, 2021.

Variables	Mean squares				
	Environments	Materials	Materials×Environments	Error	CV (%)
DTT (days)	231.0729**	76.5479**	4.6301ns	5.4078	2.77
DTS (days)	330.5104**	82.3001**	8.2075ns	9.2051	3.39
PLANTH (cm)	2283.5357**	1648.2034**	231.9838ns	247.4623	6.17
EARHT (cm)	2271.4551**	1071.4993**	167.2042ns	179.0688	9.22
TOTLEAV (No.)	28.0219**	2.3277**	0.3048ns	0.2516	3.91
LEAVA (No.)	0.8232**	0.8235**	0.0920ns	0.0769	5.68
LEAVB (No.)	19.4944**	1.2048**	0.2194ns	0.1683	5.16
LEAFLT (cm)	2012.7661**	170.2030**	28.0998ns	27.6463	6.30
LEAFWD (cm)	13.4516**	2.3407**	0.9822ns	0.9440	10.66
LAREA (cm ²)	2.63x10 ⁵ **	2.62x10 ⁴ **	5.86x10 ³ ns	5.24x10 ³	12.64
IHEIGHT	0.0121**	0.0048**	0.0010ns	0.0010	5.81
ILEAVES	0.0090**	0.0026**	0.0004ns	0.0003	4.79
PEDLT (cm)	123.5164**	11.7498**	5.8245ns	4.8536	7.40
LTBRS (cm)	10.4447**	6.5270**	2.5131ns	2.0931	17.25
LCENB (cm)	365.1503**	23.3128**	8.0770ns	7.6653	8.30
TTSLT (cm)	428.4658**	32.8915**	8.4053ns	8.8540	7.13
LBLS (cm)	569.5034**	24.7095**	11.4092ns	13.6228	15.64
PRIMB (No.)	43.2571**	9.2987**	5.4380ns	4.1409	31.52
SECB (No.)	5.3221**	0.6795*	0.4498ns	0.4438	52.83
ILTBTS	0.0082**	0.0028**	0.0013ns	0.0010	16.19
ILCBTS	0.0080**	0.0029**	0.0014ns	0.0010	4.09
ILPEDTS	0.0154*	0.0131**	0.0053ns	0.0043	9.17
ITSLPHT	0.0041**	0.0003**	0.0002ns	0.0002	8.73
EARLT (cm)	12.7784**	5.4765**	0.7387ns	0.6763	5.66
EARDM (cm)	145.7456**	37.2044**	4.6781ns	4.8240	4.63
ROWNO (No.)	3.0363*	7.7469**	1.0049ns	0.9028	6.22
KRROW (No.)	45.3528**	30.6330**	5.5470ns	5.3703	7.65
IEAR	0.0032**	0.0015**	0.0004ns	0.0004	6.51
KRLT (mm)	5.6537**	4.4132**	0.6346ns	0.4842	4.76
KRWD (mm)	0.4573*	1.0620**	0.0980ns	0.1129	4.24
KRTK (mm)	0.0175ns	0.1509**	0.0376ns	0.0578	5.88
KRVOL (mm ³)	1.66x10 ⁴ **	1.36x10 ⁴ **	1.60x10 ³ ns	1.71x10 ³	8.73
HECWT (kg hL ⁻¹)	0.4579ns	37.8905**	6.6597**	4.3825	2.96
IGRAIN	0.0304*	0.0875**	0.0123ns	0.0096	5.32
IWDLT	0.0029ns	0.0091**	0.0011ns	0.0010	5.86
ITKLT	0.0013ns	0.0043**	0.0005ns	0.0006	8.99
ITKWD	0.0007ns	0.0040**	0.0009ns	0.0010	6.38

*: $P \leq 0.05$, **: $P \leq 0.01$, ns: not significant, CV= Coefficient of variation. Degrees of freedom: environments: 2; materials: 63; interaction: 126; error: 147 (125 in ear- and kernel-related variables). The abbreviations are described in Table 2.

Four groups were identified in the dendrogram at a cut-off distance of 0.11 units (Figure 1). The first (Group IA) was formed only by the Cónico type racial controls (Cónico and Elotes cónicos), suggesting little relationship with the studied populations. Inspection of the remaining groups showed that locality of origin was not an important grouping criterion, but grain color was. Thus, Group IB was composed of 16 materials: 12 landraces, all with pigmented grain (eight with blue, two with yellow, and two with red) and four of the six experimental varieties ('Nopalucan Precoz' and 'Nopalucan Ultraprecoz', both with white grain, and the composite varieties LHM Amarillo and Azul). Group IIA included seven materials, six of which had white grain and the rest yellow grain. Among the materials with white grain were the commercial control Niebla® and a creolized population derived from this hybrid, registered as C003(WN). Finally, group IIB was the most numerous, consisting of 39 materials, dominated by white grain populations (25 landraces), followed by blue (6 landraces), yellow (2), pinto (2), and a cacahuacintle-type material registered as Z027(C). Within this group were located the experimental materials 'Serdán' and LHM Blanco, as well as the Chalqueño control, suggesting a greater resemblance of the landraces contained in this group (which represented 67 % of the total) with this race.

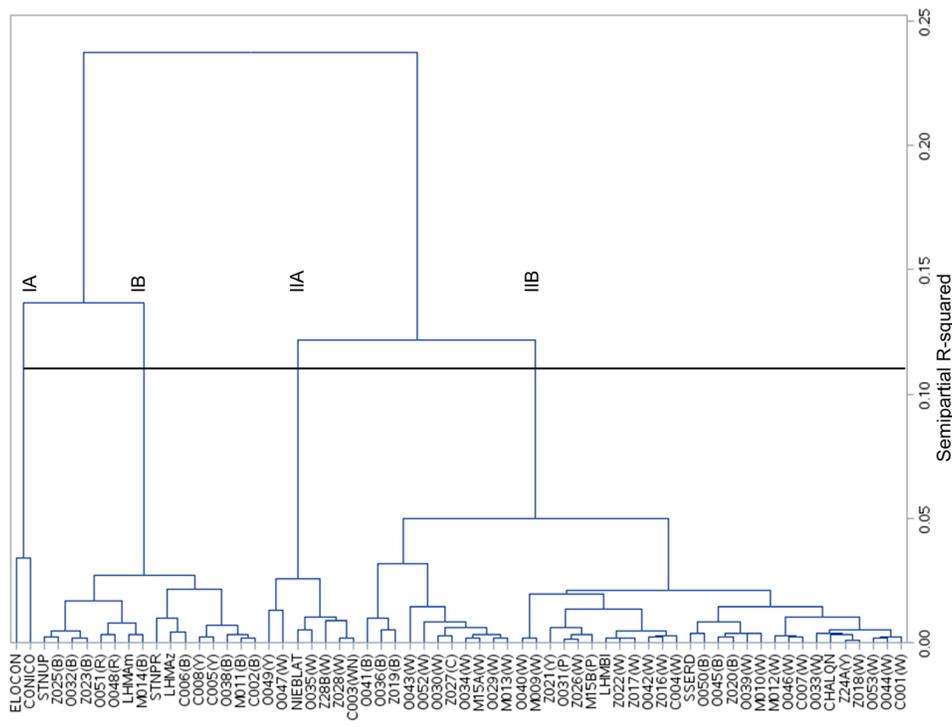


Figure 1. Dendrogram obtained with Ward's method for 54 maize landraces, three racial controls, six experimental varieties, and one commercial control evaluated in the Coronango municipality, Puebla, 2021.

In the landraces, the first letter corresponds to the locality of collection (C: Coronango, M: Mihuacán, O: Ocotlán, Z: Zoquiapan), the following positions to the accession number, and, in parentheses, the grain color (Y: Yellow; W: White; P: Pinto; R: Red; B: Blue).

The statistics associated with the multivariate analysis of variance used to compare the means of the identified groups had a probability value < 0.0001 (Table 4), thus showing that the null hypothesis was rejected and that, therefore, at least one vector of group means was statistically different from the rest.

Table 4. Test criteria and F approximations of the multivariate analysis of variance used to compare the four groups of maize materials identified in the cluster analysis.

Statistic	Value	F Value	Degrees of freedom in the model	Degrees of freedom in the model errors	Pr > F
Wilk's Lambda	0.0100	5.71	108	133.57	< 0.0001
Pillai's Trace	3.1204	4.73	108	144.00	< 0.0001
Hotelling-Lawley Trace	24.2047	7.09	108	96.63	< 0.0001
Roy's Greatest Root	14.6732	19.56	27	36.00	< 0.0001

A review of the means from identified groups in the dendrogram (Table 5) showed that the IA group was the most contrasting in comparison with the other groups, including the two Cónico type racial controls. Its plants were the shortest in height (however, their ears were inserted at a greater height than in the other groups) and had the lowest number of total leaves and leaves above the ear (coinciding with Group IB). The ear leaf area was the smallest of all groups. Their tassels were the shortest in length and had the least number of primary branches. In addition, when compared to other groups, the length of the branched part (ILTBTS index) and the central branch (ILCBTS index) represented, respectively, the smallest and the largest proportion of the total tassel length. Its ears were the shortest and narrowest, with the smallest number of rows and kernels per row, and their kernels had the smallest volume and hectoliter weight. In addition, several indices relating to kernel dimensions showed the lowest values. In general, the group included materials with small plant dimensions, which contrasted markedly with the rest of the materials under study.

IB group was the earliest of all. Regarding the other two groups that contained landraces (groups IIA and IIB), this was the one that had the plants with the lowest height, with fewer total leaves and leaves above the ear, and less ear leaf area. It showed the tassels with the shortest total and basal lateral segment lengths, and the highest ILPEDTS ratio (peduncle length/tassel total length). Its ears were shorter, with fewer kernels per row, and wider than long (IEAR = 0.30). Its hectoliter weight was the lowest among the groups with landraces (Table 5).

Compared to all groups, group IIA was characterized by showing the highest number of leaves above the ear and the highest leaf index, and by having its ears inserted at a lower height. In contrast to plants in groups IB and IIB (which also included landraces), its tassels had the lowest number of primary and secondary branches, but, in relation to the total plant height, its tassels represented the highest proportion of all groups (ITSLPHT index). Its ears were longer than wide (IEAR = 0.33) and its kernels had a low length/width ratio, but presented the highest ratios for width/length and thickness/length ratios, suggesting wider and thicker kernels than long ones. IIA was the group with the highest hectoliter weight (Table 5).

Finally, group IIB was distinguished from the rest mainly by tassel attributes: it presented the greatest length of the basal lateral segment of the tassel, the greatest number of secondary branches, and one of the greatest numbers of primary branches. The index relating total tassel length to plant height (ITSLPHT) was the lowest of all quantified (Table 5).

Groups IIA and IIB were very similar in earliness, plant height, number of total leaves, ear leaf area, peduncle length and total tassel length, and in the ILTBTS, ILCBTS, and ILPEDTS indices (which related length of the branched segment, of the central branch, and of peduncle to total tassel length, respectively), as well as in ear length and diameter, number of rows, kernels per row, kernel volume, and kernel thickness/kernel width ratio.

Principal component analysis showed that the first three components explained 66.47 % of the total variation among the materials. Principal component (CP) 1 tended to be associated with leaf area (LAREA, eigenvector = 0.30), while CP2 was associated with the number of leaves above the ear (LEAVA, 0.32), leaf index (ILEAVES, 0.34), kernel width to kernel length ratio (IWDLT, 0.31) and kernel length to kernel width ratio (IGRAIN, -0.30). Finally, CP3 was related to the number of rows (ROWNO, 0.37), and the ratios length of the central branch (ILCBTS, 0.36), and tassel branching space (ILTBTS, -0.38) with respect to total tassel length, and the number of primary tassel branches (PRIMB, -0.35).

When the groups identified in the dendrogram were represented in Figure 2, it was confirmed that there was little relationship between the Cónico type racial controls and the landraces studied, and that there was a greater association of these with the Chalqueño race control. It was also observed that groups IA and IB presented lower values of leaf area than IIA and IIB, and that considering CP2, group IIA had the highest values of leaves above the ear (LEAVA), of the proportion of these leaves with respect to the total number of leaves (ILEAVES) and of the kernel width/kernel length ratio (IWDLT), but lower values of the kernel length/kernel width ratio (IGRAIN). Finally, it is noted that within each group (and as a whole) there was an important variation in the number of rows (ROWNO), primary branches (PRIMB), and the magnitude of two indices that relate measures of tassel length (ILCBTS, ILTBTS).

Table 5. Mean test for four maize groups and 27 variables evaluated in Coronango, Puebla, 2021.

Variable	Group				HLS D
	IA	IB	IIA	IIB	
DTT (days)	90.92a	85.03b	91.86a	90.71a	4.25
PLANTH (cm)	194.00c	240.60b	248.37ab	246.76a	19.50
TOTLEAV (No.)	11.74b	12.12b	13.30a	13.08a	0.71
LEAVA (No.)	4.30c	4.57bc	5.60a	4.91b	0.37
LAREA (cm ²)	355.93c	506.79b	612.29a	604.37a	62.86
ILEAVES	0.37b	0.38b	0.42a	0.38b	0.02
IHEIGHT	0.60a	0.56b	0.52c	0.58ab	0.03
PEDLT (cm)	25.71b	30.08a	28.54a	30.07a	2.08
TTSLT (cm)	33.59c	39.31b	43.50a	42.81a	2.25
LBLS (cm)	18.95c	21.73b	24.28ab	24.46a	2.59
PRIMB (No.)	4.13b	6.31a	5.64ab	6.78a	1.97
SECB (No.)	1.18ab	1.21ab	0.79b	1.37a	0.45
ILTBTS	0.14b	0.20a	0.18a	0.21a	0.03
ILCBTS	0.86a	0.80b	0.81b	0.80b	0.03
ILPEDTS	0.78a	0.77a	0.66b	0.71b	0.06
ITSLPHT	0.18ab	0.16bc	0.18a	0.16c	0.01
EARLT (cm)	10.47c	13.62b	15.64a	14.84a	0.89
EARDM (mm)	34.86b	45.84a	47.67a	48.54a	3.22
ROWNO (No.)	13.51b	14.68ab	15.77a	15.52a	1.92
KRROW (No.)	19.73c	28.50b	31.70a	31.24a	2.34
IEAR	0.30b	0.30b	0.33a	0.31ab	0.03
KRVOL (mm ³)	331.01b	477.54a	451.21a	482.70a	76.19
IGRAIN	1.61b	1.85a	1.63b	1.90a	0.17
IWDLT	0.64a	0.55b	0.62a	0.53b	0.05
ITKLT	0.41a	0.28c	0.32b	0.27c	0.03
ITKWD	0.64a	0.52b	0.53b	0.51b	0.04
HECWT (kg hL ⁻¹)	61.44c	68.87b	73.72a	71.02ab	3.73

HLS D: Honest Least Significant Difference. Means with the same letters within rows are not statistically different (Tukey, $P \leq 0.05$).

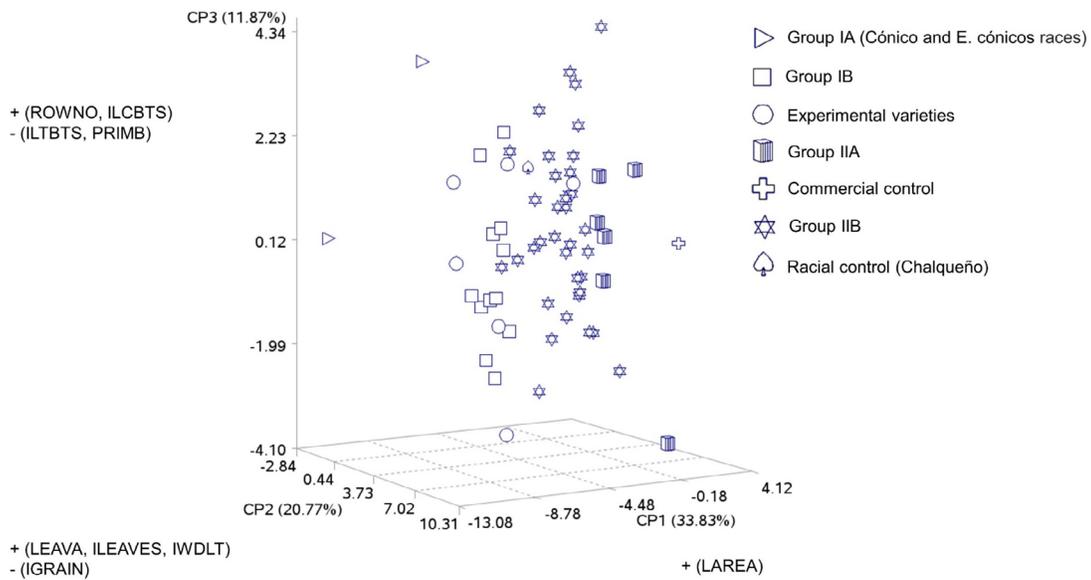


Figure 2. Dispersion of 54 maize landraces, three racial controls, six experimental varieties (three synthetics and three composite varieties) and one commercial control, based on the three main components of 27 variables evaluated in Coronango, Puebla, 2021.

Discussion

Even though Coronango municipality is small (it has an area of 3,655 ha, of which just over 2,000 ha are dedicated to agriculture (CEIGEP, 2022; SIAP, 2021) and has only one climatic type, there were differences among environments in terms of the expression level of the measured variables, with Zoquiapan being the most favorable, followed by Mihucán and Ocotlán. Such variation was caused by the fact that in the first two localities, accumulated precipitation during the crop cycle exceeded the annual average of the municipality, which could have benefited the plants since there were no severe moisture restrictions. On the other hand, Ocotlán besides having less precipitation, was exposed to significant winds occurring during the flowering stage, which lodged the plants, a situation that has been documented to affect maize development (Wang *et al.*, 2022). The non-existent interaction between materials×environments suggests that the ordering or hierarchy of the materials was maintained when passing from one environment to another, which implies that the differential response of one material, compared to another, was the same, independently of the environment considered (Cubero & Flores, 1995), a highly recommendable aspect in characterization studies (Sánchez *et al.*, 1993).

Based on the objective of this research, the main findings were the following: a) In a rural area under an urbanization process, important levels of morphological diversity among cultivated maize persist; b) Maize landraces from the study area show greater morphological similarity with the Chalqueño race.

In a rural area undergoing urbanization, there are still important levels of morphological diversity among cultivated maize.

The grain coloration pattern found in the Coronango municipality reveals that the types reported 50 years ago by Cervantes & Mejía (1984) for the Puebla Valley are maintained: white, blue, yellow, pinto and red, with the prevalence of white grain populations (70.4 % in residual moisture and 48.1 % in rainfed; 55.6 % at present) and the persistence of varieties with pigmented grain, although with a higher percentage of those of blue color at present (5.5 % in residual moisture and 12.4 % in rainfed in the collection of the years 1970-1971; 25.9 % in the present study). The later result could be attributed to a strategy that production units close to urban areas have generated, consisting of transforming the blue-colored grain into tortillas for sale (Lerner & Appendini, 2011).

Even though the study was carried out in a geographically small space (compared to more extensive studies conducted in the Puebla highlands, such as those of Hortelano *et al.*, 2008; Hortelano *et al.*, 2012 and Alvarado-Beltrán *et al.*, 2019), which is also in a process of absorption by the urban environment (Guevara, 2017), important levels of morphological variation were still detected among the landraces. Evidence of the latter is that it was possible to determine the existence of three large groups, which were differentiated from each other, particularly by grain color, earliness, and characteristics related to various plant structures (leaves, tassel, ear, and kernel). The importance of grain color and earliness as grouping attributes of landraces was documented by Muñoz (2013), who explained that within the micro-regions where maize landraces are grown, peasants have structured varietal patterns (understood as systems that combine groups of landraces, environmental strata or environmental levels and the relationships between them), whose components are defined precisely in terms of grain color and earliness. It is common for the components of the varietal pattern in maize to be represented by a set of white grain materials (late cycle) and others with pigmented grains (early cycle), as reported by López *et al.* (2020). The relevance of vegetative, tassel, ear, and kernel variables (such as leaf area, number of leaves above the ear, leaf index, width/length and length/width kernel ratios, ear row number and number of primary branches of the tassel, which contributed most to the explanation of the variation found) for group distinction has been demonstrated in previous reports, such as those of López-Romero *et al.* (2005), Vidal-Martínez *et al.* (2018) and Linares-Holguín *et al.* (2019), among others.

As in previous reports by Hortelano *et al.* (2008) and Hortelano *et al.* (2012) in micro-regions of the central highlands of Puebla, in this research, the groups of landraces were also differentiated, first by grain color and then by plant characteristics. In agreement with those studies, it was found that plants from pigmented grain materials tended to present lower values in phenological, vegetative, ear, and kernel variables than those from white grain materials.

These elements suggest that the diversity structure of rainfed maize landraces in the study area corresponds, in general terms, to that identified in larger areas of the central highlands of Puebla, where maize is grown under residual moisture.

Obtained results show that farmers in the study area have two large groups of maize materials, those with pigmented grain (Group IB) and those with white grain, with the latter subdivided into two groups, one comprised of landraces (the most numerous, Group IIB) and the other (Group IIA), which, due to its characteristics, shows affinity with improved varieties and includes a creolized material. In the case of landraces, the importance of the peasant having the two coloration groups mentioned above is that, as stated by Muñoz (2003), Castillo-Nonato (2016), and López *et al.* (2020), it enables them to face the variability in environmental conditions, managing the different levels of earliness that are associated with grain color (white, late-cycle and pigmented, early cycle) and, on the other hand, to satisfy various cultural aspects. The presence of creolized materials reflects another strategy followed by farmers to diversify their range of production options, as it results in populations that combine desirable characteristics of improved varieties with those of landraces (Bellon & Risopoulos, 2001).

The maize landraces of the study area show greater morphological similarity with the Chalqueño race.

According to Sierra-Macías *et al.* (2016), the main maize races present in the Puebla Highlands (altitude higher than 2000 m) are Cónico, Elotes Cónicos, Chalqueño, Arrocillo amarillo, and Cacahuacintle, all of them belonging to the Cónico group. Based on the collection made between 1970 and 1971 in the Puebla Valley (the present study area is contained therein), Cervantes & Mejía (1984) reported Cónico and Chalqueño as the predominant races at that time. Data obtained in the present study suggest that in the explored area, the maize landraces studied have little relationship with the Conico and Elotes Conicos racial controls (which formed a morphologically independent group) and that the greatest affinity (particularly in the case of the white grain and some pigmented materials) was with the Chalqueño racial control; therefore, it is likely that a considerable fraction of the maize currently cultivated in the area corresponds to the aforementioned race. This affinity was also corroborated by comparing the expression levels of various morphological characters recorded in this work with those reported by Wellhausen *et al.* (1951) for the Chalqueño and Cónico races; the greatest resemblance was recorded with the former.

The greater association of the maize landraces of Puebla central highlands (particularly those with white grains) with the Chalqueño race and the lesser relationship with the Cónico race is a previously reported situation by Hortelano *et al.* (2008), Hortelano *et al.* (2012) and Alvarado-Beltrán *et al.* (2019), who propose gene flow, recombination between populations, selection imposed by peasants and the greater productive capacity of the Chalqueño-type materials as the reasons to explain such association. In this work, as in those mentioned above, it was found that part of the pigmented maize did not group with the races Cónico or Elotes Cónicos, but neither was it closely related to the Chalqueño race. This may be because they are part of the existing continuum of variation in the region (Cervantes & Mejía, 1984; Hortelano *et al.*, 2008) or because they are undergoing a process of morphological differentiation that separates them from

the representative accessions of such races (Hortelano *et al.*, 2012). Another factor could be what Herrera *et al.* (2004) reported in the sense that Chalqueño is a race that shows different variation degrees and which, among other groups, includes what they called the Chalqueño-Cónico group (with intermediate characteristics between both races) and the Elotes Chalqueños-Chalqueño group, integrated by three subgroups: Chalqueño Cremoso, Chalqueño Palomo and Elotes Chalqueños (with blue grain and floury endosperm). Populations that were not grouped likely belong to one of these groups. Finally, it is possible that, in the case of the creolized materials, they exhibit features that are no longer so typical of races reported as prevalent in the region.

According to Guzzon *et al.* (2021), the reasons that lead to the abandonment of maize landraces (and, therefore, to the loss of diversity) are complex, and include agronomic, ecological, economic, and social factors, and within the latter, they cite increasing urbanization. This led to the assumption that, in the studied area, which is under strong urban growth pressure (Hernández, 2019), the existing diversity would be minimal. The results show that this was not the case, since significant morphological variation was detected, comparable to that existing in the seventies of the last century (except for races, where no samples corresponding to the Cónico race were found). This implies that, despite the pressures they face, the remaining peasants have sought to maintain the cultivated diversity, a situation that is consistent with that reported by Orozco-Ramírez & Astier (2017) in the Patzcuaro Lake Region and by Lerner & Appendini (2011) in the Toluca-Atlacomulco Valley. Notwithstanding the above, it is advisable to assess the relevance of undertaking actions aimed at conserving the diversity that still exists.

Conclusions

In the study area, which is characterized by the pressures faced by rural areas in the process of urbanization, there is still an important level of morphological diversity among the maize cultivated therein, revealed in the existence of well-defined groups that are distinguished by grain color, earliness, and plant and ear attributes. Given their characteristics, these maize landraces, particularly those with white grains, are associated with the Chalqueño race.

Authors' contribution

Work conceptualization, PTF, AGM, PAL, SESG; methodology development, PTF, AGM, PAL, SESG; software management, PTF, PAL; experimental validation, AGM, PAL; results analysis, PTF, AGM, PAL; data management, PTF, AGM; manuscript writing and preparation, PTF, AGM; writing, revising, and editing, PTF, AGM, PAL, SESG; project manager, PTF, AGM; fund acquisition, PTF, AGM.

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Conflict of interest

The authors declare that they have no conflict of interest.

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