

Mexican Maize Landraces for Corn on the Cob Production at the Central Highlands

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KeyWords *Zea mays*, tender maize, degrees Brix, sensory analysis

Abstract

Consumption of corn on the cob (*Zea mays* L.) in Mexico is based on commercial hybrids, even though there are several native races selected by farmers for this purpose. The aim of this study was to characterize immature ears from genetic material derived from three maize landraces adapted to the Mexican central highlands. We evaluated 28 accessions belonging to landraces Cacahuacintle, Chalqueño and Elotes Cónicos in Montecillo, Estado de México under a complete randomized blocks design with two replications for two consecutive cycles. Quality was measured through total soluble solid content, protein content and a sensory analysis, along with morphological traits and yield. We found native genotypes with superior quality than that of the commercial control. An analysis of variance and comparison of means by Tukey's test, as well as a principal component analysis were applied. Native maize showed soluble solid contents of 13.6° Brix, protein content of 12.4 % and better tasting of immature ears than that of the commercial control. Also, the landraces produced ear length of up to 18.5 cm, diameter of up to 5.3 cm, average of 17 grain rows and 90.8 % of grain fill length, which were similar to the control. The yield of landraces reached up to 12 t ha⁻¹ of fresh unhusked ears and 6.3 t ha⁻¹ of fresh kernels. Ear length and diameter can be used as selection criteria for improvement of yield and morphologic traits, and soluble solid content for improvement of tender corn quality. Variability in native maize could satisfy the consumption preferences of corn on the cob and fresh kernel consumption of the population centers of the central part of Mexico

Introduction

Maize (*Zea mays* L.) can be consumed as corn on the cob (tender maize ears) at the milk stage of grain-filling, with a moisture content between 72 and 76 % (Williams, 2014). This use offers advantages over dry mature grain because the fresh ears are harvested earlier, thus reducing the growing cycle, and the still green plants can be used as fresh forage for cattle feeding once the corn ears are harvested (Coutiño et al., 2010).

In the market of fresh corn ears physical traits of visual importance for consumers must be considered to develop a product of good quality. These characteristics are different through diverse markets. The market preference (Lertrat and Pulam, 2007) and the maize germplasm genetic background (Ortiz-Torres et al., 2013) have influenced the selection of maize varieties used for this purpose. In the international market it has been established that corn ears must: i) be no less than 14 cm long (Luchsinger and Camilo, 2008) and ii) have at least 16 straight rows and full grains to the corn cob apex (Shelton and Tracy, 2015). In breeding programs

aimed to the development of corn on the cob varieties, in addition to yield, quality traits are also sought after: taste, texture, sugar content and long shelf life (Lertrat and Pulam, 2007) in order to get products more agreeable and tasty (Shelton and Tracy, 2015).

Currently, in Mexico commercial hybrids, especially A-7573, prevail for producing tender ears (Valdivia-Bernal et al., 2010). Nationwide 67,697 ha were sown in 2018 for that purpose, with an average yield of 15.6 t ha⁻¹ (SIAP, 2018); the main states in production of corn on the cob were Puebla, Morelos and San Luis Potosí. In the State of Mexico 7.5 % of the total national area was planted to this crop with a yield close to the national average. Cacahuacintle, Chalqueño and Elotes Cónicos are Mexican landraces from the central highlands locally used for grain and tender maize ears, which farmers have selected in accordance to their preferences.

Other studies have evaluated maize landraces from different regions of the country for fresh corn production, and have reported ear yield, ear size (length, diameter, row number, grains per row), grain

size (length, thickness and width), soluble solids content (Ortiz-Torres et al., 2013; Coutiño et al., 2015), shelf life (Valdivia-Bernal et al., 2010) and taste (Fernández-González et al., 2014).

Considering the high human population density at Central Mexico, as it includes Mexico City and several large surrounding cities, this region has a huge requirement of fresh corn; nevertheless, there is no published scientific information on the local maize landraces used for fresh corn production, and no breeding programs focused on the production of maize varieties for fresh ears are active in the above mentioned area.

The aims of this study were: 1) to evaluate native populations of the Cacahuacintle, Chalqueño and Elotes Cónicos races in regard to traits of fresh grain quality, physical appearance and yield; and 2) to identify both parents and key traits to be used in maize breeding for fresh ears

Materials and Methods

Plant material

Twenty-eight accessions of three maize landraces from central Mexico were obtained from the International Maize and Wheat Improvement Center (CIMMYT) germplasm bank and from the active collection of the maize breeding program at Colegio de Postgraduados. More in detail, the study comprised: 10 accessions of the race Cacahuacintle, 15 of Elotes Cónicos and three of Chalqueño. The commercial hybrid A-7573 was added as control because it is considered as the reference for corn in cob production in Mexico (Table 1).

Table 1 - Maize accessions belonging to three landraces adapted to the central highlands that were evaluated for tender ear traits in Texcoco, Mexico and a commercial hybrid used as control.

Race	Accession/Trade name	Color	Endosperm type
Cacahuacintle	C-Pue-185, C-Pue-473, Zac-Q-12, Cacah-529, C-Pue-487, C-Pue-472, C-Pue-491, C-Pue-476, C-Pue-482, C-Pue-343.	White	Floury
Elotes Cónicos	Hgo-524, Hgo-503, Hgo-515, Hgo-437, Hgo-453-1, Hgo-454-1, Hgo-451-1, Hgo-513, Vino, Hgo-416, Hgo-453-2, Hgo-451-2, Pue-618, Hgo-454-2, Hgo-415.	Blue and red	Flint-like
Chalqueño	Mich-195, Mex-491, Hgo-428.	Yellowish white	Flint
Commercial control	A-7573.	White	Flint

Experimental site

Plants were field grown during the Spring-Summer cycle for two consecutive years (2015 and 2016) in Texcoco, State of Mexico, Mexico (19° 29' N, 98° 53' W, at 2250 masl). The climate type of this site is Cb (wo) (w)(i) g, which is temperate with long warm summers, mean annual temperature between 12 and 18 °C and mean annual rainfall of 637 mm (García, 1998).

Experimental design and unit

In both years, the experiments were conducted under a randomized complete blocks experimental design with two replications. The experimental unit was a plot of six 5-m long rows and 0.8 m apart, containing 22 useful plants at a density of 55,000 plants ha⁻¹. Plots were supplied of water by irrigation, and fertilized with 180N-60P-0K applied at two growth stages: at sowing with half the nitrogen dosage (urea) and all the phosphorus (ammonium sulfate); the rest of the nitrogen was applied 45 days after sowing.

Morphological traits and yield of fresh corn ears

The harvest of fresh corn was done 20-21 days after female flowering as optimal stage for the tender kernel consumption as corn on the cob (milk-dough stage) (Shelton and Tracy, 2015; Trimble et al., 2016); therefore, the plots were harvested at different dates, depending on the flowering date of each genotype. The flowering date was recorded when 50 % of the plants showed exposed silks.

In each plot five unhusked ears were harvested from plants with complete competition; then, length (LE, cm), diameter (DM, cm), grain row number (RN) and length of filling from the base to the apex (LF, %) was registered for each ear. The yields of unhusked corn (FY, t ha⁻¹) and of fresh kernel (FKY, t ha⁻¹) were estimated from the measured data as indicated above. In order to standardize FY and FKY data, the kernel moisture content was adjusted to 70 % (Rice and Tracy, 2013; Williams, 2014) and to a plant density of 50 thousand ears ha⁻¹ (potentially marketable ears).

Fresh corn quality traits

Total soluble solids content (TSS, °Brix). The extract obtained from a uniform mixture of grains prepared from 25 ears in consumption stage was evaluated for this trait. The ear harvest was performed early in the morning before sunrise, and the ears were kept in ice until their processing. The TSS determination was done in triplicate with a digital refractometer (Atago Pal-1®, Tokio, Japan) in 300 µL of extract.

Total protein content (TP). It was measured in a uniform mixture of 100 g of fresh tender kernels removed with a knife from 25 ears; then, the sample was dried in an oven at 70° C during 72 h until constant weight. The dried grains were grinded in a cyclonic mill during 3 min, and three sub-samples were obtained to determine the moisture content in an oven at 105 °C for 24 h; this moisture was used to adjust the final nitrogen content (on dry matter basis). The protein content was determined with the Micro-Kjeldahl 46-13.01 method (AACC, 1999).

Kernel taste (KT). This trait was determined through a

extension. Since not all the traits met the assumption of normality, the Spearman's correlation coefficients were calculated in order to identify linear relations between pairs of variables. In addition, a canonical correlation analysis between quality and morphological traits and yield was done. The two analyses of correlation were done with standardized data to normality with mean equal to 0 and variance equal to 1, in order to equalize the measurement scales between traits. Additionally, a principal component analysis was performed based on the correlation matrix to determine the descriptive value of each trait to the observed variability and

Table 2 - Mean squares from the analysis of variance corresponding to the evaluation of 28 maize accessions from three landraces for fresh corn traits. Montecillo, State of Mexico, 2015 and 2016.

Source of variation	Quality			Visual appearance				Yield	
	TSS [‡]	TP	KT	LE	LF	DM	RN	FY	FKY
Genotypes (G)	2.9*	2.9***	0.6**	9.1***	17.4 ns	775.8***	14.0***	14.1***	3.3***
Years (Y)	9.4*	12.8**	0.4 ns	23.0***	18.4 ns	1527.9***	0.1 ns	139.3***	86.9***
G × Y	1.3 ns	1.4 ns	0.3 ns	1.7*	13.6 ns	144.9 ns	0.7 ns	2.1 ns	1.4*
Replicates/ Y	8.5**	0.6 ns	0.2 ns	0.8 ns	0.4 ns	14.0 ns	1.1 ns	1.9 ns	1.1 ns
Error	1.5	0.8	0.3	0.9	9.1	92.0	0.5	1.3	0.8
CV (%) [†]	11.6	8.2	8.1	5.4	3.5	6.3	5.4	12.5	17.3

*, **, ***Significant with $P \leq 0.05$, $P \leq 0.01$ y $P \leq 0.001$, respectively; ns, Not significant; [‡]TSS, total soluble solids; TP, total protein; KT, kernel taste; LE, length; LF, length of grain filling; DM, diameter; RN, grain row number; FY, fresh corn yield; FKY, fresh grain yield; [†]CV, coefficient of variation.

sensory test done with an untrained panel of 40 people (women and men from three age groups, < 30, 30-50 and > 50 years old). The cooked corn was prepared using 1 kg of fresh kernels in the stage for consumption (Mexican "esquite") of each genotype and then it was cooked with no additives in potable water during 1 h on a portable electric stove. Thereafter, portions of 30 g of cooked corn were presented to the panel at typical consumption temperature (≈ 45 °C) in a small plastic cup (32 mL). The order for presenting the samples to each member of the panel was randomized, and the palate was cleansed with potable water between samples. The taste evaluation was based on a 9-point hedonic scale (1: I extremely dislike it, 9: I extremely like it) as proposed by Stone et al. (2012).

Statistical analyses

A combined analysis of variance across years was carried out for each trait, and the assumption of normality was verified. The ear diameter did not adjust to a normal distribution, not even with data transformation, so a non-parametric test was applied by the assignment of ranks combined with the Friedman test (Friedman, 1937). The mean comparison was done with the Tukey test ($P \leq 0.05$), complete data is presented as additional information due to its

use this information to define selection criteria for plant breeding. All data analyses were done with the statistical software SAS 8.0 (SAS Institute, 1999).

Results and discussion

Genotypes, traits, years: analysis of variance

The analysis of variance showed significant differences among genotypes ($P \leq 0.05$) for all the evaluated traits, except for length of the grain filling (Table 2). The effect of years was significant for all the measured traits, except for grain taste and grain rows in the ear. It is interesting that consumers perceived differences between genotypes, but not between the two years (environments) of evaluation, even though other quality traits varied significantly between years. The interaction G × Y had no effect over quality of corn, but it significantly affected the ear physical appearance (LE) and ear fresh yield (FY), although the interaction effect was usually much lower than the main effects G and Y.

Results show that the studied maize accessions have promising genetic variability for developing commercial varieties for corn on the cob production with genetic background of maize landraces. The new varieties would be aimed to produce fresh ears and fresh kernel (Mexican *esquites*) in the environmental conditions

of the Mexican central highlands, where the tested varieties, except the hybrid control, are broadly used for producing both mature grain and fresh ears

Fresh corn quality traits

Mexican maize landraces of better ear quality than the commercial control were identified in this survey, since they have higher content of total soluble solids (TSS) in the fresh kernels and total protein (TP), as well as flavor (KT). TSS is an indicator of the sugar content in the corn kernels (Favarato et al., 2016), although TSS also includes organic acids, vitamins, phenolic compounds, pectin, etc. All the native maize accessions evaluated contained more TSS (11-13.6 °Brix) than the commercial hybrid used as control (9.7 ° Brix) (Figure 1); that is, these Mexican accessions are sweeter than the control. One accession of the Chalqueño race (Mich-195) reached 13.6 °Brix, while Coutiño et al. (2010) reported a higher value (15.2 °Brix) in the ‘Campechano’ variety that belongs to the Tuxpeño race in the Southern Mexican state of Chiapas. According to Coutiño et al. (2015), in most Mexican markets the ear sweetness does not strongly determine the consumer preference, as it does happen in the US,

Canada, India and other Asian countries (Singh et al., 2014). Sweetness in corn kernels can reach up to 24-30 °Brix in the fresh stage as corn on the cob, thus classifying as a quality product for the food processing industry (Luchsinger and Camilo, 2008).

Regarding the nutritional aspect, accessions of the maize races Chalqueño and Elotes Cónicos showed the highest average protein content (12 %), that surpassed the control and other landrace maize groups which averaged 10.3 % (Vidal et al., 2008) and 11.9 % (Vera-Guzmán et al., 2012); both research teams found that these protein contents are similar to that of the high quality protein maize, but are different in their proportions of essential amino acids such as lysine and tryptophan (Vidal et al., 2008).

As for the fresh corn taste (KT), this trait should be evaluated at the market consumption stage (Lertrat and Pulam, 2007). In this study the best taste rank was attained by accession C-Pue-472 that belongs to the Cacahuacintle race, a position significantly above the control hybrid, as it was graded by our sensory evaluation panel, thus, setting a perspective for the Mexican consumption preference in the central region of the country. The lowest KT grades (~ 5.5) were

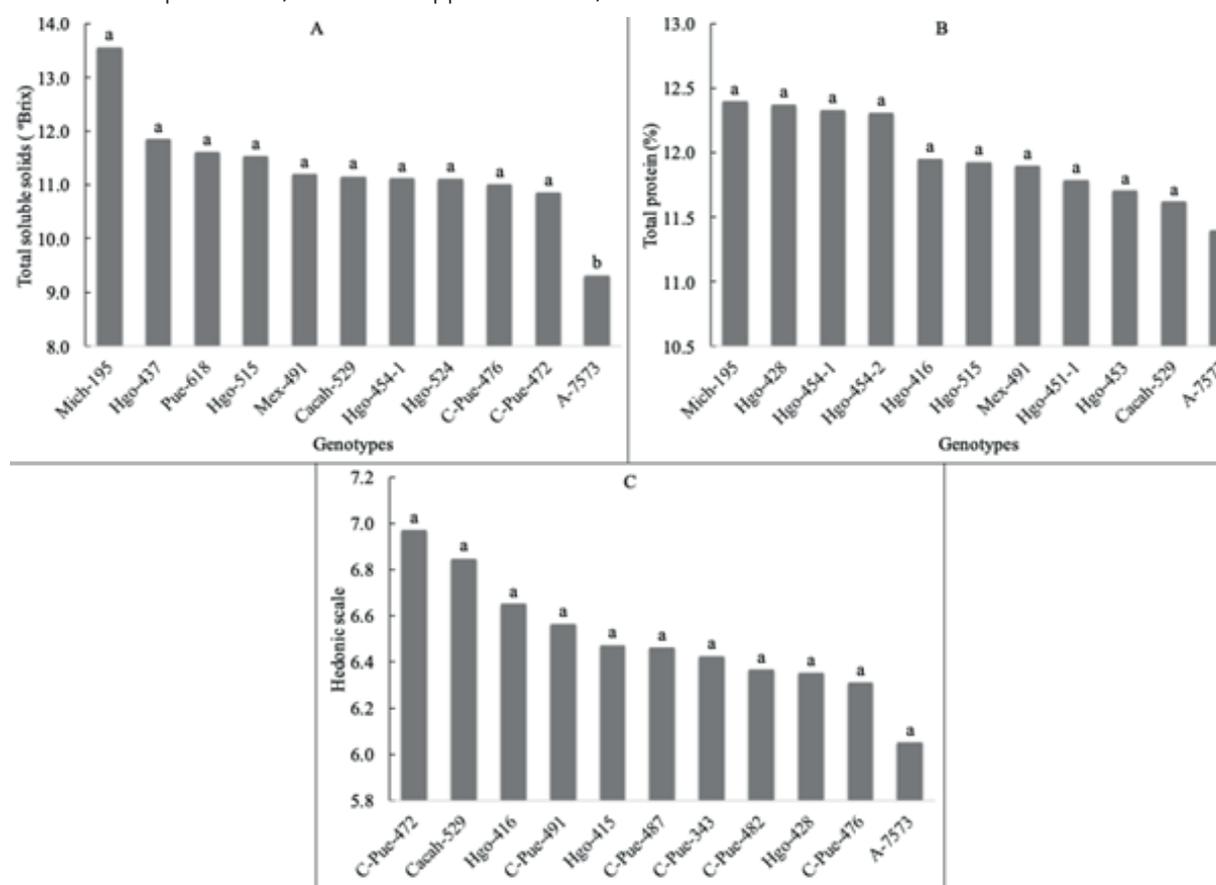


Fig. 1 - Outstanding maize landraces and the commercial control for fresh corn quality traits: A) Total soluble solids content; B) Protein content; C) Fresh kernel taste

Table 3 - Spearman's correlation coefficients between fresh corn traits in 28 maize landraces adapted to Mexican highlands, 2015 and 2016.

Traits	TSS	TP	KT	LE	LF	DM	RN	FY	FKY
TP	0.11 ns	1.00							
KT	-0.13 ns	-0.05 ns	1.00						
LE	-0.28*	-0.06 ns	0.34**	1.00					
LF	-0.07 ns	0.01 ns	0.01 ns	0.41**	1.00				
DM	-0.26*	-0.35**	0.14 ns	0.35**	-0.06 ns	1.00			
RN	0.15 ns	0.15 ns	-0.55***	-0.43***	-0.18 ns	0.14 ns	1.00		
FY	-0.25*	-0.32*	0.14 ns	0.60***	0.13 ns	0.80***	0.11 ns	1.00	
YKY	-0.33*	-0.31*	0.07 ns	0.48***	0.20 ns	0.65***	0.19 ns	0.88***	1.00

*, **, ***Significant at $P \leq 0.05$, $P \leq 0.01$ and $P \leq 0.001$, respectively; † ns, not significant; ‡TSS, total soluble solids; TP, total protein; KT, kernel taste; LE, length; LF, grain-fill; DM, diameter; RN, kernel row number; FY, fresh corn yield; FKY, fresh kernel yield.

associated with the blue colored kernels of the Elotes Cónicos maize race. Even though the blue corn kernels are locally very well appreciated when prepared as 'tortillas', for fresh kernels ('esquites') this colored grains caused curiosity among the panelists who compared them to black beans (*Phaseolus vulgaris* L.), a staple food in the Mexican cuisine. Nevertheless, for the state of Chiapas in Southern Mexico, Coutiño et al. (2015) claim that farmers and housewives prefer corn varieties with black, red or yellow fresh kernels, as they are associated with better taste and sweetness in relation

to the white grains; thus, the market preferences of corn on the cob greatly vary among regions across the country. In our study both women and men preferred the white fresh grains. Half of the participants with less than 30 years of age preferred white grains (71.4 %), while panelists between 30-50 of age and those older than 50 years favored the yellowish white grains (43.8 and 57.1 %, respectively). No one preferred the blue fresh kernels.

This study highlighted maize accessions with comparable kernel quality to that of other maize

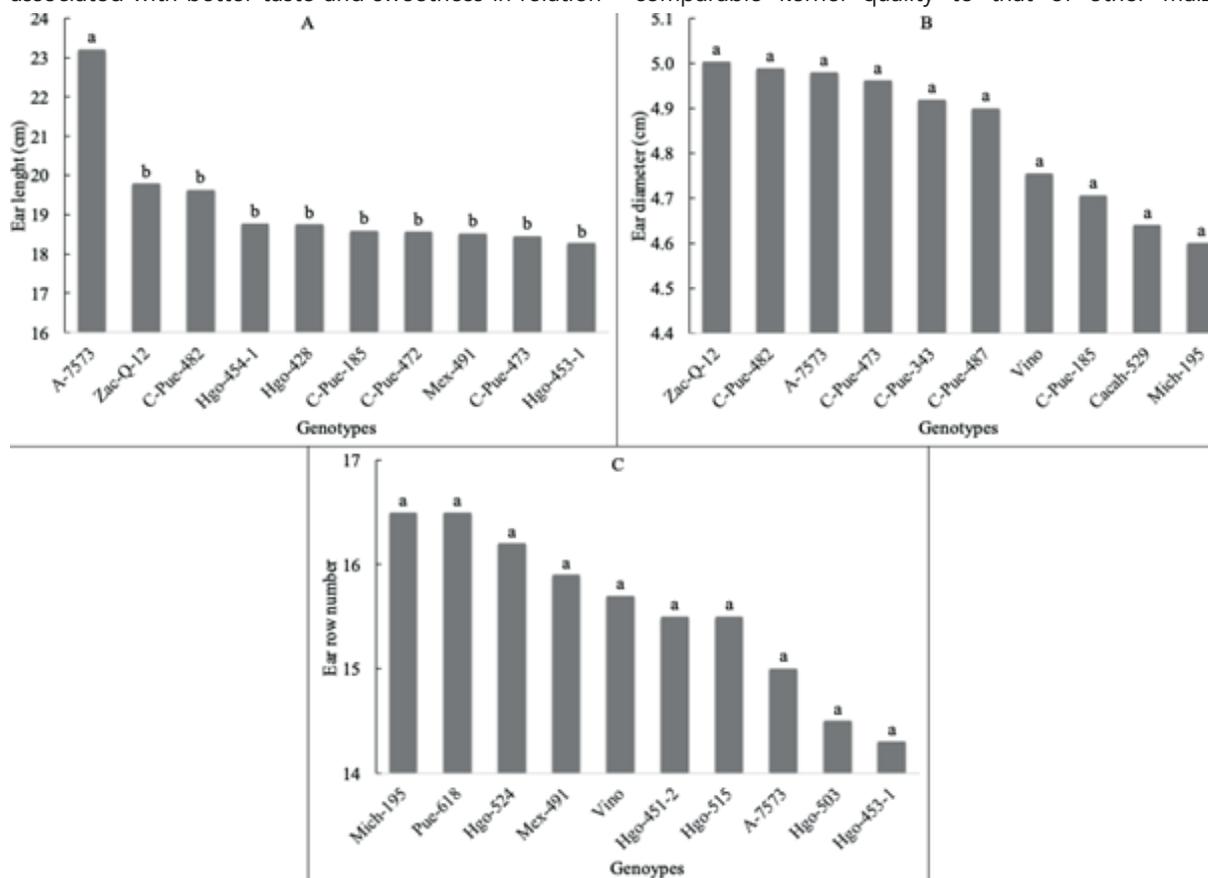


Fig. 2 - Outstanding maize landraces for morphological maize ear traits: A) Length; B) Diameter; C) Kernel row number.

landraces or commercial varieties evaluated in other studies; kernel sweetness and flavor, and protein content of the Mexican landraces are the quality traits that will distinguish corn varieties derived from the native tender ears in the market. In addition, kernel sweetness can be improved by introgression of other Mexican sweet corn races like Dulce de Jalisco or Dulcillo del Noroeste, which might also increase the preferences and the environmental adaptability

Fresh corn morphological characteristics

Genotypes showed significant differences in physical appearance of the ear (Figure 2), except for length of grain-fill which showed a mean coverage value of 86.4 %. Among native maize accessions ear length varied between 15.5 (Elotes Cónicos) and 19.8 cm (Cacahuacintle), diameter fluctuated from 3.8 (Elotes Cónicos) to 5.1 cm (Cacahuacintle), and showed at least 16 kernel rows in the Chalqueño and Elotes Cónicos races. In some markets, length and diameter are the main characteristics that fresh corn consumers most look for (Coutiño et al., 2015); while in others consumers prefer rows to be straight and having grains all the way up to the ear tip (Shelton and Tracy, 2015). Among the evaluated genotypes, we found in this study different ear sizes that could meet various markets or personal preferences. Even though ear length resulted higher for the hybrid control than for landrace accessions, we believe it still will not determine the consumer preference if the new improved maize varieties have an ear length similar to that of some accessions as landrace Zac-Q-12, by virtue of a better flavor and quality traits

Fresh yield

Retail sales of fresh corn on the cob is done by piece, so that physical ear aspect and size must satisfy each target market, while for *esquites* sales, only the quantity of fresh kernels obtained from the ear is considered. The accessions derived from landrace Cacahuacintle and Elotes Cónicos resulted the best for yield of corn on the cob and fresh kernel (Figure 3), excepted hybrid control performance.

Yield of fresh corn on the cob (FY) in ZAC-Q-12 was 12.0 t ha⁻¹, 4.1 t (25.5%) less than the control A-7573 (16.1 t ha⁻¹). The highest fresh kernel yield (FKY) was registered in the control (8.3 t ha⁻¹) and the accessions C-Pue-473 (6.3 t ha⁻¹), Zac-Q-12 (6.2 t ha⁻¹) and C-Pue-482 (6.1 t ha⁻¹), which yielded significantly higher than the other genotypes (Figure 3). A reliable identification of the best native germplasm on yield traits is instrumental for the application of a well-focused breeding program in order to reach and even overcome the commercial

control. The breeding program needs to focus on corn ear quality and appearance traits such as cob length and diameter, aimed to achieve a competitive role in the market.

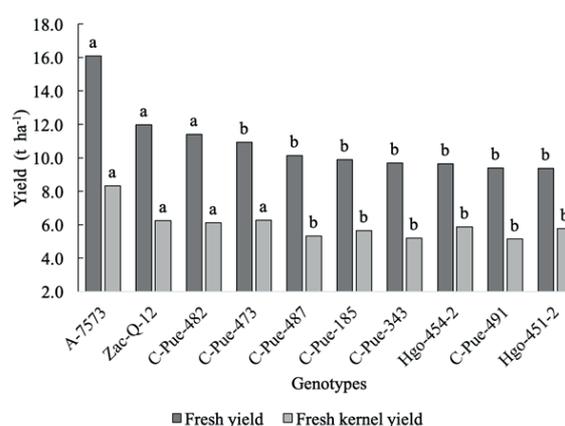


Fig. 3 - Best ten maize landraces for fresh corn and kernel yield. Means with same letters within each type of bar are not statistically different (Tukey, $P \leq 0.05$).

Fresh kernel yield is important for the food processing industry (Luchsinger and Camilo, 2008; Williams, 2014), in which sweet corn varieties dominate canned and frozen products. Non-sweet corn varieties, such as the ones evaluated in this study, are associated with the production of *esquites*. In Mexican maize landraces FKY has not been measured directly; however, it is expected that a greater FKY should be associated with lower cob proportion, and thus to a better quality and acceptance by the market. Nevertheless, Williams (2014) pointed out that it is necessary to measure directly processor variable like FKY to have an adequately prediction recovery of fresh kernel mass.

Landraces with acceptable yield of fresh corn were found, mainly those from the race Cacahuacintle. Fresh kernel yield estimation in this study offer a good alternative to select the best genotypes for production of *esquites*, the second most important way in which fresh corn is consumed in Mexico. With a proper selection breeding program, that include selection for fresh yield and ear appearance traits, it is expected that maize landraces could match and even surpass the commercial control and be competitive in the market.

Relationship between fresh corn traits

The canonical correlation analysis revealed that the group of quality traits is closely related to the group of morphological and yield traits ($r = 0.72^{***}$); such strong association can be used in breeding programs in the region for the direct and indirect selection of genotypes. Significant ($P \leq 0.05$) simple linear associations were also found between fresh corn traits (Table 3). Negative

correlations were identified among quality (TSS and TP) and morphological (DM) and yield (FY and FKY) traits.

Kernel taste (KT) was not associated with TSS, this confirms that preference of consumers represented by evaluating panel of this study is not related with sweetness, which was also noted by Fernández-González et al. (2014), but it is not in agreement with findings of Coutiño et al. (2010) and Gere et al. (2014). Yield of fresh corn and fresh kernel was positively associated ($P \leq 0.001$) with LE and DM; however, a negative association with TSS and TP ($P \leq 0.05$) was also found, which hinders simultaneous breeding of both groups of traits, although it could likely be overcome by applying appropriate selection indices. Correlations found between corn traits can be used to improve the selection process through indirect selection for specific traits, and thus get an improved variety with several desired traits.

Table 4 - Eigenvectors and eigenvalues of the first three principal components derived from six traits of fresh corn attributes of 28 maize landraces from Mexican highlands, 2015 and 2016.

Original trait	PC 1	PC 2	PC 3
TSS [†]	-0.26	0.22	0.10
TP	-0.20	0.02	0.67
KT	0.10	-0.53	-0.18
LE	0.44	-0.21	0.26
LF	0.21	-0.27	0.53
DM	0.39	0.23	-0.33
RN	0.00	0.65	0.18
FY	0.51	0.15	0.06
FKY	0.49	0.22	0.10
Eigenvalue	3.43	1.87	1.19
Variance explained	0.38	0.21	0.13
Cumulative variance	0.38	0.59	0.72

[†]TSS, total soluble solids; TP, total protein; KT, kernel taste; LE, length; LF, grain-fill; DM, diameter; RN, kernel row number; FY, fresh corn yield; FKY, fresh kernel yield; PC: principal component.

Trait relative relevance

A genotype with good agronomic performance does not always express the best quality, fresh corn yield or morphological characteristics. In this study the correlated group of traits which is considered of greater importance is formed by TSS, TP, LE, DM, FY and FKY, as it includes the three main categories of traits for corn on the cob attributes. In accordance with the principal component analysis (Table 4) from the aforementioned trait group, the first three components explained 72 % of the observed phenotypic variation.

The most relevant traits as for their involvement in the eigenvectors were FY, FKY, LE and DM. In the maize

breeding process, the data collection of FY and FKY is time-consuming and imply a great effort; therefore, LE and DM represent a good alternative to indirectly select genotypes, since they involve less effort than that required to estimate yields; in addition, these traits would be helpful to simultaneously improve for fresh corn characteristics, because they are also correlated with the latter, as shown in the Table 3.

Results imply that selecting for yield, there would have a slightly negative effect on TSS and TP, as observed in the matrix of correlations (-0.25* and -0.31*, respectively). It is advisable to supplement the use of correlations and principal components with selection indices, in order to improve quality and yield characters at the same time.

Once the best traits for obtaining improved maize landraces of corn on the cob had been identified, the next step is to establish a plant breeding program for each pertinent native maize race or by integrating the desired traits into a single genotype. Consumer preference in central Mexico is determinant for the assembly of new corn varieties of corn on the cob.

Conclusions

Maize landraces genotypes, characterized by the best quality traits, with yield and morphological traits close to those of the reference commercial hybrid for corn on the cob production in Mexico. Genotypes corresponding to the Cacahuacintle and Elotes Cónicos races were identified to have the potential to meet requirements for consumption of highly populated areas of Mexico; therefore, they become candidates as parental materials for breeding and be improved until reaching competitiveness, with the advantage of having better quality traits like higher content of total soluble solids and better kernel flavor.

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