

Physicochemical characteristics and quality of the protein of pigmented native maize from Morelos in two years of cultivation

Elizabeth Broa Rojas¹
María Gricelda Vázquez Carrillo^{2§}
Néstor Gabriel Estrella Chulím¹
José Hilario Hernández Salgado¹
Benito Ramírez Valverde¹
Gregorio Bahena Delgado³

¹Postgraduate College-Campus Puebla. Boulevard Forjadores de Puebla núm. 205, Santiago Momoxpan, San Pedro Cholula, Puebla. CP. 72760. Tel. 01 (222) 2851442. (broa.elizabeth@colpos.mx; nestrela@colpos.mx; jhhernan@colpos.mx; bramirez@colpos.mx). ²Valle de México Experimental Field-INIFAP. Highway Los Reyes-Textcoco km 13.5, Coatlinchán, Textcoco, State of Mexico. CP. 56230. Tel. 01 (800) 088 2222, ext. 85364. ³School of Higher Studies of Xalostoc-UAEM. Av. Nicolás Bravo s/n, Industrial Park, Cuautla, Morelos. CP. 62725. Tel. 01 (777) 3297981. (gbahena20@yahoo.com.mx).

§Corresponding author: gricelda.vazquez@yahoo.com.

Abstract

Historically in east Morelos state, pigmented native maize (MNP) has been produced and consumed, so it was proposed to evaluate the effect of the crop cycle on the physicochemical characteristics and protein quality of 26 MNP populations of the municipalities of Temoac and Ayala of the eastern region of Morelos. The MNP were collected in the spring-summer 2014 and 2015 cycles, which were classified as: hectoliter weight (PH), weight of one hundred grains (PCG), flotation index (IF), pedicel (PED), pericarp (PER), germ (GER), flourey endosperm (EHA) and corneal (ECO), as well as oil (ACE), protein (PRO), tryptophan (TRI), protein quality index (IQP) and anthocyanins (ANT). The results were analyzed with a completely randomized combined design. In the eastern of Morelos, the Western Elotes 'EO' and Pepitilla 'Pep' of blue grain predominated. In all the variables significant differences were found ($p < 0.01$) in the culture cycle, the population and the interaction. In the 2014 cycle, the maizes were higher: PCG, ECO, ACE, TRI and IQP and lower: IF, PRO and ANT. In general, the MNP were of low density ($\bar{X}_{PH} = 63.7$ kg hL⁻¹; $\bar{X}_{IF} = 84$), large grain ($\bar{X}_{PCG} = 44.8$ g), with high: GER ($\bar{X} = 13.1\%$), ACE ($\bar{X} = 5.14\%$), TRI ($\bar{X} = 0.076\%$), IQP ($\bar{X} = 0.81$) and ANT ($\bar{X} = 276.6$ mg ECG kg⁻¹ MS). The populations 'EO22' and 'Pep20' stood out due to their high IQP, high TRI and ANT, the rest of the populations showed no relationship between chemical and nutritional characteristics. It is identified 16 populations with good protein quality (IQP > 0.8) and high TRI content (>0.075%), which can be used to improve nutrition and health of consumers.

Keywords: flotation index, protein quality index, tryptophan.

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Introduction

In Mexico, corn is considered the most important crop, since almost seven million hectares are planted, of which about 23 million tons are obtained. It is estimated that 80% of the arable land is worked under rainfed conditions, where the distribution and volume of water depends on rainfall, which significantly reduces the average yield (2.3 t ha^{-1}), especially when compared to the irrigation (7.3 t ha^{-1}), (SIAP-SAGARPA, 2017). The 65% of the surface cultivated in Mexico under temporary conditions is planted with native maize (CIMMYT, 2014), which have a wide range of adaptation to very specific agroclimatic conditions and also, for its culinary characteristics such as color, flavor, texture are very appreciated by consumers for their use in the preparation of several typical dishes (Hellin *et al.*, 2013).

Such is the case of pigmented corn (blue, black, red, purple, etc.), which are planted largely to satisfy the palate of the families that produce them and to praise the guests at special events of each community. The native maizes or also called by the maize producers 'creoles' have an ancestral rootedness in the life of the Mexicans, in spite of this, the use of these maizes has been modified over the years due to factors such as globalization, changes in the social and productive life of the rural sector, changes in consumer preferences and migration (López-Torrez *et al.*, 2016).

In the state of Morelos, corn is the second most important crop after sorghum. The area planted with corn in Morelos for 2015 was 23 922 ha, where an average yield of 2.27 t ha^{-1} was obtained (SIAP-SAGARPA, 2017). The production of this cereal is mainly in temporary conditions using improved genotypes, which have gradually displaced the pigmented native maizes which are currently planted on a small scale and used mainly for self-consumption, they are used to make tortillas, carts (food made with dough and lard, cooked in a comal), pinole (prehispanic food based on roasted corn, ground and sprinkled with sugar and cinnamon), tamales (product made with dough mixed with butter which is wrapped and sewed in the bracts of the cobs 'totomoxtle') and atole (drink made from roasted and ground corn or with nixtamalized pigmented corn dough).

The aforementioned changes are present in the municipalities of Ayala and Temoac, Morelos, since in the sixties the main crop was corn and only native maize was grown. In the last thirty years these maizes were gradually replaced by improved varieties and hybrids of corn and crops such as sorghum, amaranth and peanut, in addition, in 2016 by yellow maize for animal consumption. In addition to this, the family production units have been reduced due to the advanced age of the producers, the migration of young people and the lack of interest in perpetuating their cultivation by the children who have stayed, or the grandchildren since they consider it an arduous and unprofitable activity (Barkin and Suárez, 1995).

In Mexico, the physical, chemical and structural components of the grain of pigmented native maize have been evaluated (Salinas *et al.*, 2013b), anthocyanins have also been quantified (Espinosa *et al.*, 2009; Salinas *et al.*, 2013a), as well as protein quality (Vidal *et al.*, 2008). The food industries look for raw materials with very specific rheological properties of their starches, aspects that have been studied in native maize with the purpose of positioning them in the demanding industries (Gaytan-Martínez *et al.*, 2013).

The physical-chemical characteristics and the structural components of the grain of corn depend on the variety, the conditions of the crop, as well as the methods of selection implemented by the producers, can also be modified by the climatological changes. Vázquez-Carrillo *et al.* (2016) reported that with rainfall greater than 500 mm and average temperatures of 15-26 °C in the stage of grain filling, grain size, hardness and starch content were increased.

The difference between genotypes of the same breed is explained by several factors: their intrinsic genetics, whether it is a pure race, or a crossbreed and each producer's control of pests and diseases and the management implemented in response to the weather, frost, hail, lodging, etc. The above was studied under the genotype x environment interaction analysis. In this regard, Bazinger *et al.* (2012) indicated that genotype x environment interaction occurs when genotypes respond differently to environmental variants. While Ángeles-Gaspar *et al.* (2010) reported that the genetic variation of maize is related to the factors associated with soil moisture, temperature, altitude and the duration of the plant growth period.

Therefore, the objective of this research was to evaluate the effect of the crop cycle on the physico-chemical characteristics and protein quality of 26 populations of pigmented native maize from the municipalities of Temoac and Ayala of the eastern region of the state of Morelos, Mexico.

Materials and methods

Germplasm studied

In the spring-summer (SS) -2014 cycle, 12 producers from the municipality of Temoac and five from Ayala were selected. In the SS-2015 cycle, the same producers provided between 20 and 25 ears each year (Table 1). In Ayala the climate is warm sub-humid and Temoac is temperate sub-humid. Based on the characteristics of the cobs, the race and the cross to which they belong were identified.

Table 1. Race and localities of collection of pigmented native maize from the eastern of Morelos, Mexico.

| No. of population MNP | Race ¹ (identification) | Color | Location | Municipality | Altitude (m) |
|-----------------------|------------------------------------|-------|-----------|--------------|--------------|
| 1 | ES x TAB | Red | Popotlan | Temoac | 1 619 |
| 2 | Pep x EO | Red | Temoac | Temoac | 1 599 |
| 9 | EO x Pep | Red | Popotlan | Temoac | 1 616 |
| 10 | EO | Red | Amilcingo | Temoac | 1 466 |
| 11 | EC x ES | Red | Amilcingo | Temoac | 1 466 |
| 12 | EO | Red | Amilcingo | Temoac | 1 466 |
| 13 | EO x Pep | Red | Amilcingo | Temoac | 1 466 |
| 17 | Pep x EO | Red | Huazulco | Temoac | 1 513 |
| 19 | Pep x EO | Red | Temoac | Temoac | 1 528 |
| 21 | Ni | Red | Tlayecac | Ayala | Ni |
| 23 | EO x Pep | Red | Temoac | Temoac | 1 562 |
| 3 | EO | Blue | Popotlan | Temoac | 1 560 |

| No. of population MNP | Race ¹ (identification) | Color | Location | Municipality | Altitude (m) |
|-----------------------|------------------------------------|-------|-------------|--------------|--------------|
| 4 | AN x EO | Blue | Popotlan | Temoac | 1 587 |
| 5 | Pep | Blue | Popotlan | Temoac | 1 635 |
| 6 | EO | Blue | Popotlan | Temoac | 1 618 |
| 7 | Pep | Blue | Popotlan | Temoac | 1 619 |
| 8 | Pep x EO | Blue | Popotlan | Temoac | 1 616 |
| 14 | EC x Pep | Blue | Tlayecac | Ayala | Ni |
| 15 | EC x Pep | Blue | Amilcingo | Temoac | 1 466 |
| 16 | Pep | Blue | Amilcingo | Temoac | 1 486 |
| 18 | AN | Blue | Tlayecac | Ayala | Ni |
| 20 | Pep | Blue | Tlayecac | Ayala | 1 374 |
| 22 | EO | Blue | Tlayecac | Ayala | Ni |
| 24 | Ni | Blue | Temoac | Temoac | 1 528 |
| 25 | Pep x EO | Blue | Huitzililla | Ayala | Ni |
| 26 | Pep x AN | Blue | Amilcingo | Temoac | 1 481 |

MNP= pigmented native maizes; ¹ES= Elotero de Sinaloa; TAB= Tabloncillo; Pep= Pepitilla; EO= Western Elotes; EC= Conical Elotes; AN= Ancho; Ni= not identified. Hernández-Casillas (Com. Per.).

Physical and chemical characteristics of the grain

The hectoliter weight (PH), weight of one hundred grains (PCG), flotation index (IF) and percentages of pedicel (PED), pericarp (PER), germ (GER), mealy endosperm (EHA) and corneal (ECO) was evaluated according to the methodology described by Salinas and Vázquez (2006). The chemical analysis was performed on whole grain flour obtained after milling in a cyclonic type mill (UDY, Mod.3010-080P[®]), with 0.5 mm mesh. The content of oil was quantified following the method 920.85 of the AOAC (2000), protein by the Technicon Instruments method (Galicía *et al.*, 2012), tryptophan following the glyoxylic acid methodology (Nurit *et al.*, 2009) and the protein quality index (IQP) was calculated with the relationship proposed by Twumasi-Afriyie *et al.* (2016): $IQP = [(tryptophan (\%)/protein (\%))100]$.

The total anthocyanins were evaluated following the procedure described by Salinas *et al.* (2013a). The anthocyanins of the red MNP were calculated with a chlorinated pelargonidin curve (Castañeda-Sánchez, 2011) and are reported as mg equivalents of chlorinated pelargonidin per kilogram of dry matter (mg EPC kg⁻¹ MS). For blue MNP a cyanidin-3-glucoside curve was used, reported as mg equivalents of cyanidin-3-glucoside per kg dry matter (mg ECG kg⁻¹ MS) (Salinas *et al.*, 2013a).

Statistical analysis

The data corresponding to each of the variables evaluated in each production cycle were subjected to a variance analysis under a completely randomized design combined with two replications using the GLM procedure and the Tukey comparison test ($\alpha = 0.05$). The Pearson correlation matrix was made. The statistical package SAS (Statistical Analysis System 9.0 for Windows) was used for all analyzes.

Results and discussion

In the municipalities of Ayala and Temoac located to the eastern of Morelos predominated as pure breeds: Western Elotes ‘EO’ and Pepitilla ‘Pep’, both were large grain and soft endosperm. The populations of ‘EO’ (five) had lower: PCG (\bar{x} = 42.1 g) and index of flotation (\bar{x} = 76), with respect to the populations of ‘Pep’ (four), which presented the characteristic hook of this breed, higher PCG (\bar{x} = 45.1 g) and IF (\bar{x} = 87) (Figure 1). Vázquez *et al.* (2010) reported a lower PCG (\bar{x} = 33.1 g) for the Pepitilla breed.



Figure 1. Appearance of the pericarp, germ and endosperm of a red (A) and blue (B) population of pigmented native maize from the eastern of Morelos.

The pure race of ‘Ancho’ corn (AN) was of lower PCG (\bar{x} = 37.9 g) and higher IF (\bar{x} = 99). The most frequent crosses (eight) were between ‘PepxEO’, in six of them ‘Pep’ was the primary race; however, the size and hardness of ‘EO’ PCG (\bar{x} = 42.4 g) and IF (\bar{x} = 85) predominated, while the ‘EOxPep’ crosses registered characteristics similar to ‘Pure Pepitilla’ with average PCG and IF values. of 47.7 g and 81 respectively. The crosses of ‘AN’ occurred with ‘Pep’ and ‘EO’, they were blue and very soft endosperm. Two of these populations could not be classified (Table 1), possibly due to increased cross-linking.

Wellhausen *et al.* (1951) reported that the race ‘Pep’ was distributed mainly in the states of Guerrero and Morelos and that it was of ‘soft endosperm, white grains with aleurone and pericarp without color’. In this research we found pure populations of ‘Pep’ of blue-black color of soft endosperm and with pigment in the aleurone layer. While the red ones, they had the pigment in the pericarp and some also in the aleurone layer.

In the state of Morelos seven collections of wild relatives and a good number of collections of representative creoles of eight races have been identified, for which reason it has been declared with an average variability and part of the center of origin of the corn (Gómez *et al.*, 2015). Additionally, in the 2008-2009 collection carried out in the warm zone of the state of Morelos, 14 breeds were identified, among them pepitilla and Western Elotes which were white (50-60%) and in smaller proportion (10-15%) yellow, blue and red (Gómez *et al.*, 2015). In the study municipalities, white grain maize was identified, but the research focused on the pigmented ones, among which were identified the races Elotes de Sinaloa ‘ES’ and Tabloncillo ‘TAB’ that had not been previously reported.

Physical and chemical characteristics of the grains

The analysis of variance showed a significant difference ($p < 0.01$) in the effects of crop cycle, population and the interaction between cycle x population in all the variables evaluated (Table 2).

Table 2. Mean squares and significance for physical, chemical and protein quality characteristics of pigmented native maizes from the eastern of Morelos.

| Variables | Cycle | Population | Interaction cycle*population | Error | CV | General average |
|---------------------------|----------|------------|---------------------------------|--------|------|--------------------|
| Hectoliter weight | 43.9** | 51.1** | 33.7** | 101.3 | 1.58 | 63.7 |
| Weight one hundred grains | 935.1** | 82.2** | 49.6** | 0.73 | 1.91 | 44.8 |
| Flotation index | 775.5** | 489.1** | 257.4** | 12.4 | 4.19 | 84 |
| Pedicle | 0.38** | 0.11** | 0.08** | 0.0006 | 1.42 | 1.8 |
| Pericarp | 0.22** | 0.77** | 0.41** | 0.0007 | 0.58 | 4.5 |
| Germ | 1.74** | 3.7** | 1.59** | 0.0006 | 0.2 | 13 |
| Flourish endosperm | 598.7** | 438.8** | 101.5** | 0.0007 | 0.06 | 50.5 |
| Corneal endosperm | 720.1** | 429.6** | 97.2** | 0.0007 | 0.09 | 30.2 |
| Oil | 1.39** | 0.72** | 0.35** | 0.033 | 3.58 | 5.1 |
| Protein | 4.8** | 3.8** | 2.93** | 0.08 | 2.95 | 9.6 |
| Tryptophan | 0.0003** | 0.00019** | 0.00009** | 0 | 0.91 | 0.075 |
| Quality index | 0.2** | 0.04** | 0.042** | 0.0009 | 3.73 | 0.8 |
| Anthocyanins | 456.8** | 122255.1** | 12349.3** | 1.68 | 0.47 | 276. |

** = significant $p < 0.01$; CV = coefficient of variation.

In the two years of evaluation, the maizes were of large grains, corresponding to high values of PCG, reduced PH and high IF (Table 3).

Table 3. Effect of the crop cycle on physical, chemical and protein quality characteristics in MNP from the eastern of Morelos, Mexico, SS-2014 and SS-2015.

| Variables | Years | | DMS |
|---|----------|----------|--------|
| | 2014 | 2015 | |
| Hectoliter weight (kg hL ⁻¹) | 63.07 b | 64.37 a | 0.5286 |
| Weight of one hundred grains (g) | 47.84 a | 41.84 b | 0.4497 |
| Flotation index | 81 b | 87 a | 1.85 |
| Pedicle (%) | 1.76 b | 1.88 a | 0.0136 |
| Pericarp (%) | 4.48 b | 4.57 a | 0.014 |
| Germ (%) | 12.88 b | 13.14 a | 0.0136 |
| Flourish endosperm (%) | 48.05 b | 52.85 a | 0.0146 |
| Corneal endosperm (%) | 32.81 a | 27.55 b | 0.014 |
| Oil (%) | 5.25 a | 5.01 b | 0.072 |
| Protein (%) | 9.37 b | 9.8 a | 0.1486 |
| Tryptophan (%) | 0.078 a | 0.075 b | 0.0004 |
| Quality index | 0.858 a | 0.771 b | 0.016 |
| Anthocyanins (mg ECG kg ⁻¹ MS) | 274.49 b | 278.68 a | 0.6815 |

Values with the same letter in the same line are not statistically different (Tukey, 0.05). DMS = minimum significant difference.

The cycle x population interaction showed that the blue MNP were endosperm smoother than the red ones (Figure 2A and B) highlighted the populations ‘EOxPep9’ (Figure 2A), ‘AN18’, ‘EO3’, ‘PepxEO25’, ‘PepxAN26’ and ‘PepxEO5’ (Figure 2B), for the stability in the smoothness of its grain in the two evaluation cycles and for having been the softest grain, characteristic that was associated with PH < 62 kg hL⁻¹, PCG > 38 g and reduced percentage of PER (< 4.5%). Desirable attributes in the corn intended for consumption as a vegetable (corn) (Revilla and Ordaz, 2016) and for the preparation of pozole, where the use of corn with a PH < 67 kg is recommended. hL⁻¹, PCG > 38 g, IF = 100 and pericarp < 6% (Vázquez *et al.*, 2016).

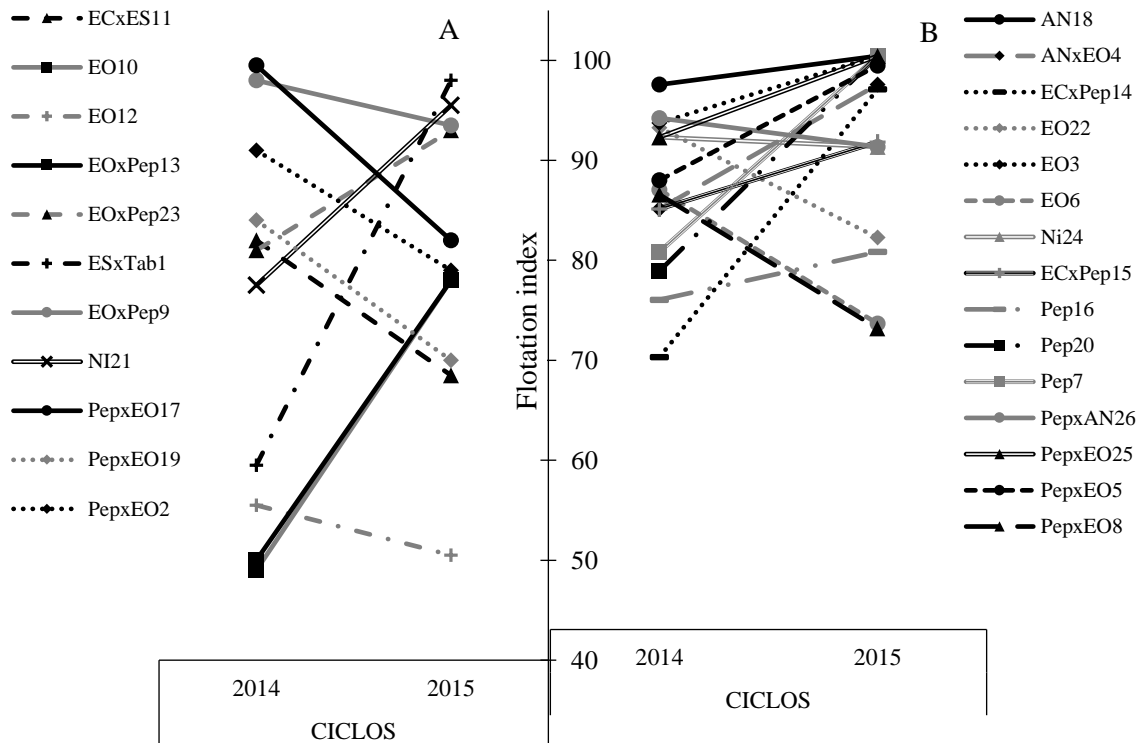


Figure 2. Interaction of crop cycle by population in the flotation index of red (A) and blue (B) populations of pigmented native maize from the eastern of Morelos, Mexico.

The greater size of the grains in the SS-2014, are attributed to a greater availability of water at the beginning of the crop and during the filling of grain (Figure 3), which favors a rapid accumulation of carbohydrates (Tanaka and Yamaguchi, 2014).

While the smaller size (<PCG) and IF, in the SS-2015 cycle are attributed to erratic and poorly distributed rain at the stage of grain filling (Ángeles-Gaspar *et al.*, 2010; Bazinger *et al.*, 2012) (Figure 3), in this regard, Zinselmeir *et al.* (1995) noted that, in corn, low water potentials reduce embryonic growth, by decreasing the flow of sucrose and altering the metabolism of carbohydrates in the ovaries. The larger grain size was associated with a large germ, which in turn produced a greater amount of oil ($r = 0.68, p < 0.01$) (Figure 4). The populations of red color, where the race ‘EO’ predominated, were those with the highest percentage of germ and oil, with respect to the blue ones (Figure 4).

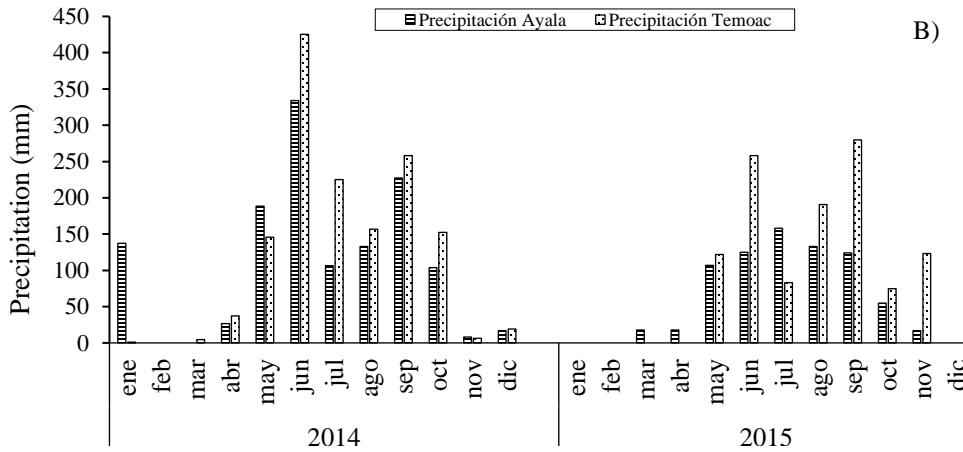
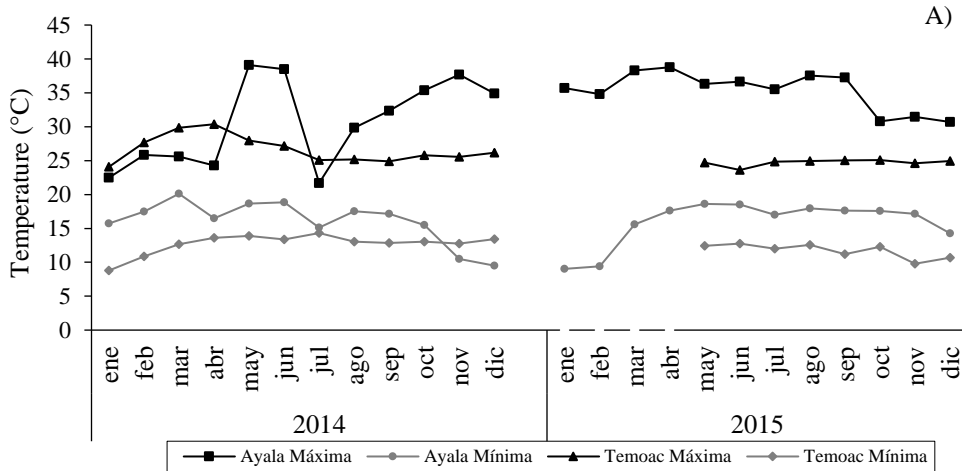


Figure 3. Temperature (A) and precipitation (B) in Temoac and Ayala, Morelos in the years 2014 and 2015 (CONAGUA, 2016).

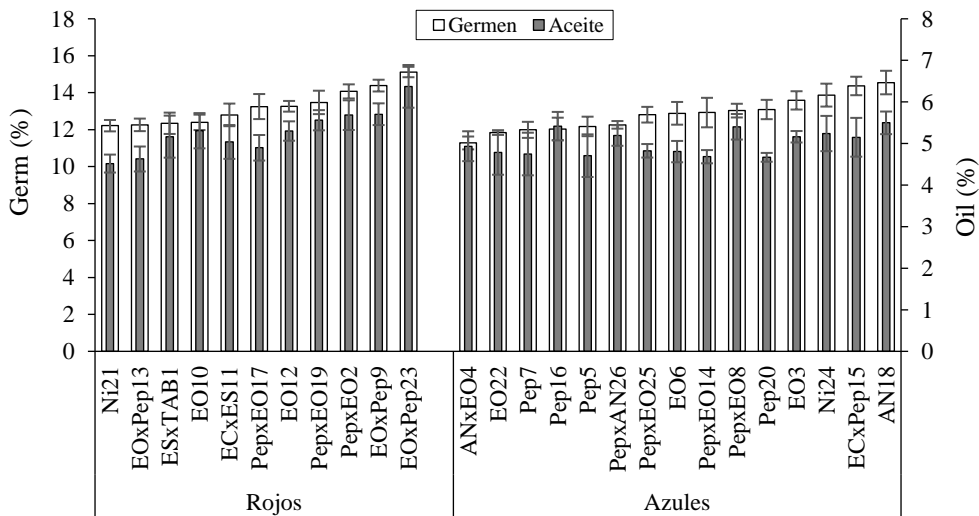


Figure 4. Oil content and proportion of germ in pigmented native maize from the state of Morelos, Mexico.

Reduced PH values were correlated with high values of: IF ($r = -0.54$; $p \leq 0.01$) and mealy endosperm (EHA) ($r = -0.61$; $p \leq 0.01$), which corroborates the low density of these maize. In its chemical components, the populations of the PV-2014 cycle recorded higher amounts of ACE and less: PRO and ANT (Table 3).

The ACE values were between 4.46 and 5.88%, with an average value of 5.14%, similar results were reported by Agama-Acevedo *et al.* (2011) (4.85 to 5.98%) for 15 accessions of blue corn and were higher than what was declared for blue (4.1%) and red (4.2%) maize of Nayarit (Vidal *et al.*, 2008) and 50 accessions of Cuban maize (4.01 to 5.53%) (Martínez *et al.*, 2009), but inferior to that found by Guzmán *et al.* (2015) in native maize of Guanajuato.

In the MNP, the protein was between 7.6 and 11.5%, values that are within what was reported for Mexican native maize (Gaytan-Martínez *et al.* (2013), these authors reported an interval between 9.7 and 11.9%, while Vázquez *et al.* (2010) reported values from 6.9 to 12.5% of protein. In the MNP of Morelos it was observed that those with the lowest protein content were those with the milder grains, where the ‘starch granules’ that are mainly spherical are weakly packaged in a protein matrix that slightly surrounds the granules (Narvaez-González *et al.*, 2007).

Another aspect that contributed to the protein content was the fertilization of the crop, where the producers reported that this occurred out of time, due to the absence of rain and in a reduced dose, added empirically (1 fist per bush). Some producers reported having incorporated the stubble from the previous crop and a small number of them, applied organic fertilizer from their draft animals. The practice of monoculture, the erratic rains and the limited application of fertilizer, are some of the causes that may have contributed to the low protein content of the populations investigated.

The crop cycle affected the content of total anthocyanins in 2015 was recorded a higher content than that of 2014 (Table 3); nevertheless, the mean values were numerically very similar in the two cycles, which could be attributed to the heritable character of the anthocyanins (Halbwirth *et al.*, 2002). The red grain populations had values from 24.79 to 623.2 mg EPC kg⁻¹ MS, with an average value of 151.4 mg EPC kg⁻¹ MS (Figure 5A).

This variability was related to the color of the grains (results not shown), those with lighter shades (rose-lilac) were those with the lowest content of anthocyanins, while the population ‘EOxPep13’ (intense red or cherry) registered the higher content of these pigments (Figure 5A). The anthocyanins of the red populations of Morelos were superior to the values reported (62.3 and 154 mg ECG kg⁻¹ MS) for unidentified maize (red) breeds (López *et al.*, 2009) and similar to that reported by Salinas-Moreno *et al.* (2012b) (64.7 to 547.7 mg ECG kg⁻¹ MS) for four races (Olotillo, Tehua, Tuxpeño and Vandeño) of red grain from the state of Chiapas. The populations of blue grain registered a higher content of anthocyanins, with respect to the red ones (Figure 5A and 5B). In the 2015 cycle, the following populations stood out: ‘Pep20’, ‘Pep16’, ‘EO6’, ‘PepxEO25’ due to its higher content of anthocyanins, while ‘EO22’, significantly reduced these pigments (Figure 5B).

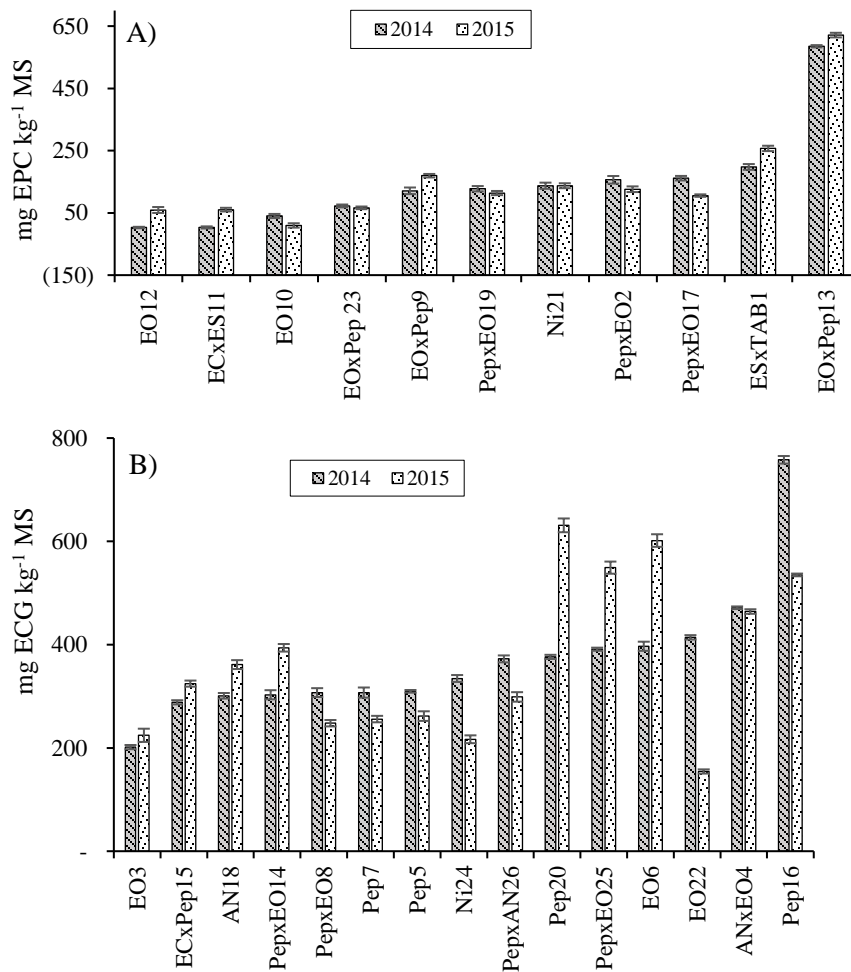


Figure 5. Total anthocyanins in populations of red (A) and blue (B) maize in the eastern region of the state of Morelos in the 2014 and 2015 cycles.

In these populations the anthocyanins had between 312 and 646.4 mg ECG.kg⁻¹ MS, with an average value of 368.4 mg ECG kg⁻¹ MS (Figure 5B), results that were similar to those reported by Salinas *et al.* (2013a) for different breeds of blue native maizes, and lower than the range of: 579.4-1046.1 mg ECG kg⁻¹ MS reported by Salinas-Moreno *et al.* (2012a) for six accessions of Chalqueño corn, a breed adapted to heights above 2 000 masl, where low nocturnal temperatures and high luminosity prevail, which favors a greater synthesis of anthocyanins (Jin-Seng *et al.*, 2006; Salinas *et al.*, 2013a). Although the genetics of the populations is important in the production of the anthocyanins, the hot sub-humid climates for the municipality of Ayala and temperate sub-humid for Temoac, could also have contributed.

Quality of the protein

The nutritional quality of corn as a food is determined mainly by the amino acid composition of its proteins. Tryptophan is one of the essential and limiting amino acids of corn (Vázquez *et al.*, 2012). Although there are no absolute values of lysine or tryptophan that define a corn with protein quality, Twumasi-Afiyie *et al.* (2016) suggested that maize with more than 0.075%

tryptophan in whole grain samples, can be considered with protein quality. In the present study, the highest tryptophan content was found in the SS-2014 cycle (Table 2). Values were recorded from 0.064 to 0.09%, with an average value of 0.075%. According to Twumasi-Afiyie *et al.* (2016), 62% of the populations (16 of 26) had a tryptophan content similar to that of maize with high protein quality (QPM) (Figure 6).

The blue populations with the highest tryptophan content were: ‘PepxAN26’ (0.090%), ‘AN18’ (0.089%), ‘ECxPep15’ (0.085%), followed by pure populations of ‘Pep7’ and ‘EO22’, while the red ones were; ‘PepxEO19’ (0.086%) and ‘EOxPep9’ (0.081%) (Figure 6).

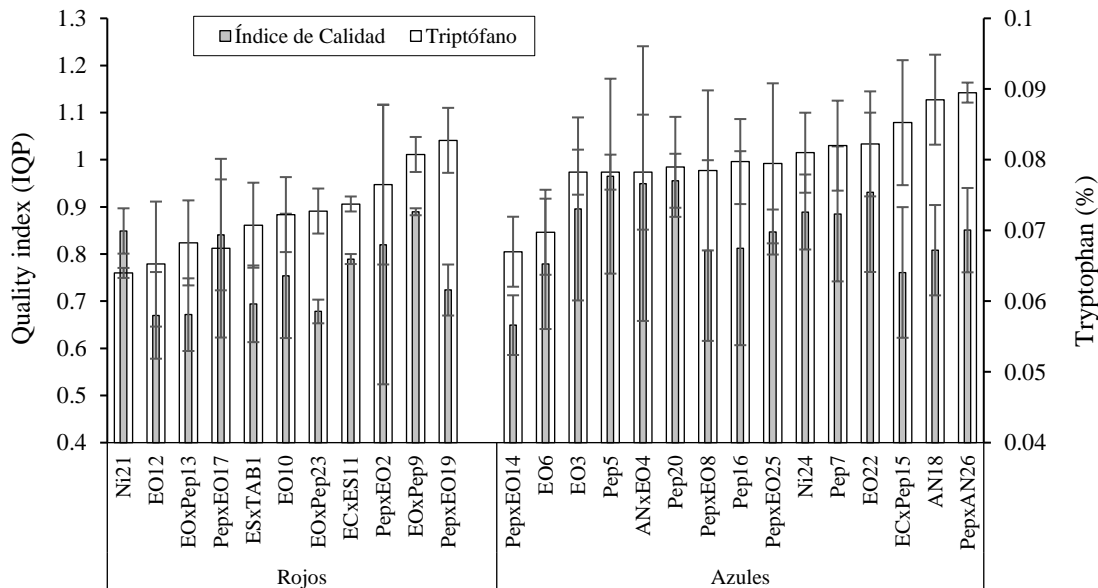


Figure 6. Tryptophan and protein quality index in populations of pigmented native maize from the state of Morelos, Mexico.

The populations with less tryptophan were ‘Ni21’, ‘ECxES11’, ‘ESxTAB1’, ‘EO10’ (red) and ‘PepxEO14’ (blue), this variability shows that tryptophan is a component that can be modified by the genetic information of each population (Krivanek *et al.*, 2007), but also due to environmental conditions and agronomic management (Vázquez *et al.*, 2012). The tryptophan content of the eastern populations of the state of Morelos was higher than that reported by Vidal *et al.* (2008) (0.048 and 0.051%) and lower than the results of Martínez *et al.* (2009) who found an interval from 0.053 to 0.097%.

It has been reported that the contents of tryptophan and lysine in corn are highly correlated (>0.9), so, with the quantification of tryptophan, it is possible to qualify the quality of the protein as an indicator of its nutritional value (Nurit *et al.*, 2009). Additionally, one of the most used criteria to identify a corn with protein quality is the protein quality index (IQP). In the MNP, the IQP presented values higher than 0.8, indicated as the limit to consider a corn with protein quality (Twumasi-Afiyie *et al.*, 2016), on the other hand, Martínez *et al.* (2009) proposed that the selection of maize with nutritional attributes should include the values of protein, tryptophan and IQP of: 8%, 0.075% and 0.8% respectively.

In the 2014 cycle, a higher tryptophan content was obtained (Table 3); however, in 2015 the populations ‘AN18’ (\bar{x} = 0.093%), ‘ECxPep15’ (\bar{x} = 0.092%) and ‘PepxAN26’ (\bar{x} = 0.091%) were those with the highest TRI. In the general evaluation (the two study cycles), it shows that 90% of MNP populations had protein content above 8%. In the case of tryptophan, 15 populations were above 0.075% (Figure 6).

Finally, 61% (15/26) of the populations registered an IQP \geq 0.8 (Figure 6). According to these three variables, populations stood out; ‘Pep5’, ‘Pep20’, ‘EO22’ and ‘ANxE04’ all blue (Table 4). They were characterized by having very soft, large endosperm grains, with higher oil and tryptophan content and less protein (Table 4) than that of a commercial toothed corn (\bar{x} = 9%) (Watson, 2003).

Table 4. Nutritional quality and physicochemical characteristics of outstanding populations of pigmented native maize from the eastern of Morelos.

| Population | Flotation index ^Ω | Weight of 100 grains | Oil [£] | Protein [£] | Tryptophan ^{£, ¥} | Protein quality index ^π |
|------------|------------------------------|----------------------|------------------|----------------------|----------------------------|------------------------------------|
| | | (g) | (%) | | | |
| ANxE04 | 91 MS | 52.6 | 4.94 | 8.52 | 0.078 | 0.95 |
| EO22 | 87 MS | 48 | 4.78 | 8.9 | 0.082 | 0.93 |
| Pep5 | 93 MS | 42.7 | 4.71 | 8.54 | 0.078 | 0.97 |
| Pep20 | 89 MS | 44.8 | 4.78 | 8.31 | 0.079 | 0.96 |

^ΩNMX-032 (2002); MS= very soft; [£]reported values on a dry basis; [¥]= in whole grain sample; ^πIQP= [(tryptophan%/protein%)100].

Thus, the maize with the best protein quality, were not those with the highest protein, nor those with more tryptophan, but those that balance the values of these three variables (Table 4). MNP can be used to improve the nutritional conditions of rural families in conditions of malnutrition. The identified diversity can be explained by the changes that the local races have experienced over time, derived from the crossing with other races, as a result of the selection of the producers, which has been directed mainly to maintain the color and the special flavor that present this type of corn with respect to improved varieties and commercial hybrids. MNP from the eastern state of Morelos are a good option to produce them under variable climate conditions and to elaborate value-added foods and can contribute to address nutrition and health problems.

Conclusions

In the pigmented native maize of Morelos, the pure blue races predominated: Western Elotes and Pepitilla, combined with each other or with other breeds such as: Ancho, Elotes Cónicos, Elotes de Sinaloa, Tabloncillo and two unidentified. These populations were of reduced density, with soft or very soft endosperm, large grain, with high percentage of germ and oil and low pericarp and protein. The blue MNP had more anthocyanins than the red ones, the blue ‘Pep16’ population standing out because of its high content of this pigment.

It is identified 16 populations with good protein quality (IQP > 0.8) and high tryptophan content (>0.075%), which can be used to improve the nutrition and health of consumers. The crop cycle associated with the climatic variations affected the physicochemical characteristics and the quality index of the pigmented native maizes. In 2015, extreme drought and increased temperature increased the grain's softness and germ ratio, but reduced grain size, tryptophan content and protein quality index.

This evidenced that, faced with a possible scenario of increased temperature and lack of rain due to climate change, these pigmented native maizes can thrive.

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