

RESEARCH ARTICLE

Morphological characterization of wild populations of *Solanum lycopersicum* var. *cerasiforme* in the tomato domestication area

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ABSTRACT

Wild tomatoes *Solanum lycopersicum* var. *cerasiforme* are found in regions with wide environmental diversity, in the state of Veracruz, México, which has originated morphological variations within this group leading to his adaptation to different environments. The aim of this study was to investigate the morphological variations of *S. l.* var. *cerasiforme* in his domestication area. We collected plants and fruits during the field trips, and we extracted seeds from these fruits to sow them later, under the greenhouse. We evaluated 35 characters according to the IPGRI (International Plant Genetic Resources Institute) descriptors in 10 collections of *S. l.* var. *cerasiforme*, we used a completely random design. In order to compare de collections and analyze the variables we performed variance analysis, principal component analysis and cluster analysis with euclidean distance (UPGMA clustering method). We found significant differences between the variables (Tukey, $P \leq 0.05$), except for the style length and flower number per plant variables. We found that 91 % of the collections have a pistil inserted and 9 % exert. The first two components explained 78 % of the total variation. The dispersion of the collections in the four quadrants of the first two components indicated great phenotypic diversity. We found four groups and we observed the greatest variation in the yield and reproductive variables. This study shows intraspecific morphological differences between wild tomato populations in the state of Veracruz, a state with a wide environmental and ecosystem diversity. It can be said that in terms of morphological characterization of wild tomato species and cultivated tomatoes for both the pattern of variability is the same. The populations we have studied could constitute a valuable germplasm bank for genetic improvement programs.

Keywords: Phenotypic variation; Germplasm; Genetic diversity; Wild tomatoes

INTRODUCTION

Tomato is one of the most popular and important vegetables in the world (Salim et al., 2018). The world production exceeds 180 million tons (Mata-Nicolás et al., 2020). It is an annual self-pollination plant belonging to the Solanaceae family with chromosome number $2n = 2x = 24$. The *Lycopersicon* clade contains the cultivated tomato *Solanum lycopersicum* and 12 wild relatives (Peralta et al., 2005, 2008). It is widely accepted that tomatoes took its origin in the Andean region between Bolivia, Chile, Colombia, Ecuador and Peru (Peralta et al., 2005; Blanca et al., 2012). The evidence of diverse types and forms of tomato found in Mexico suggest that its

domestication took place in Mexico particularly in Puebla and Veracruz (Bai and Lindhout, 2007).

Wild tomato *Solanum lycopersicum* var. *cerasiforme* migrated from Peru to Mesoamerica as a spontaneous plant by natural means, and in Mexico exists environmental, ecological conditions favorable to their development and domestication (Jenkins, 1948; Rick, 1974). In Mexico before the arrival of the Spanish, large tomato fruits were encountered, produced for human consumption, which is an artificial selection index. Based on linguistic and cultural evidence, Bauchet and Causse (2012) coincide with Jenkins (1984) on the place of domestication of the tomato, so that the word tomato took its origin from Nahuatl (Aztec) (Saavedra et al., 2017), the natives of Mexico

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called the tomato “xitomatl”. Based on the molecular and morphological evidence, *S. l. var. cerasiforme* is the direct progenitor of cultivated tomato and the domestication process is likely a two-step process, which started with the pre-domestication of *S. l. var. cerasiforme* from the wild tomato *Solanum pimpinellifolium* followed by migration of *cerasiforme* to Mesoamerica, where the true domestication occurred and finally led to the generation of the cultivated tomato bearing big fruits (Ranc et al., 2008; Blanca et al., 2012; Liu et al., 2020). The Mexicans from XIV to XVI century cultivated tomato in a polyculture system called milpa (González Carmona and Torres Valladares, 2014), and actually tomato crop has low genetic and morphologic variation within and between cultivars due to its narrow genetic base (García-Martínez et al., 2006; Bai and Lindhout, 2007). For this reason, wild tomato ancestors are important, they provide several genes capable of giving crops resistance to factors that cause abiotic as well as biotic stress; they play an extremely important role in genetic improvement programs (Foolad, 2007; Bergougnoux, 2014). According to the various explorations carried out, the wild form *S. l. var. cerasiforme* grows in diverse environments from the subtropics and semi-dry regions in the Mexican territory, from Sinaloa (Sánchez-Peña et al., 2006) to the Yucatan peninsula. In Veracruz, Delices et al. (2019) observed *S. l. var. cerasiforme* growing in different regions with different ecosystems, ranging from mesic to extremely arid conditions. Despite this wide distribution, there is lack of information about their genetic, morphological diversity. It is known that plants can modify their phenology, morphology, physiology and development in response to the environmental conditions in which they are found (Fischer et al., 2011). This ability to adapt to different environments leads to morphological, physiological, and phenotypic changes (Fischer et al. 2011), which are directly related to gene expression (West-Eberhard, 2008). Morphological traits are useful for genetic evaluation (Stoilova and Pereira, 2013), crop yield and reproductive value (Agong et al., 2001; Dharmatti et al., 2001; Mohanty et al., 2001; Parthasarathy and Aswath, 2002). Restrepo et al. (2012), studied the diversity within the *Solanum* genus and found great variation in the traits, fruit weight, fruit size, day to flowering, number of seeds per fruit. Restrepo et al. (2006) found that the variables (days to flowering and stem diameter) explained 76.3% of the total variation, in wild germplasm of (*Lycopersicon* spp). Scintu et al. (2015), evaluated a wide collection of tomato (*Solanum lycopersicum* L.) for morpho-phenological, quality and resistance traits, they reported wide variation for traits like number of flowers per inflorescence, fruit weight and number of locules; they found high coefficient of variation (CV) of 36.28, 73.77 and 62.41% respectively for these variables. One of the characteristics reported by Blanca et al. (2012) in her work was that many features of the wild tomato are based on their geographical location; reported as an example the position of the style that varies from inserted, slightly exerted,

completely exerted. These authors highlighted great diversity between accessions mostly in variables such as growth type, inflorescence type, fruit color, fruit cross-sectional shape. Wild germplasm has been studied in order to know about their potential (Flores-Hernández et al., 2017; Marín-montes et al., 2016), to find important traits of interest for breeding (Crisanto-Juárez, 2010; Carrillo-Rodríguez et al., 2010) and to take conservation measure (González-Aguilera et al., 2011). The aim of this study was to assess the morphological and phenotypic diversity of tomato accessions from wild populations *S. l. var. cerasiforme* collected in several regions (north, center and south) of the state of Veracruz, Mexico, based on 35 tomato descriptors from International Plant Genetic Resources Institute (IPGRI).

MATERIALS AND METHODS

We collect plants and fruits of wild tomatoes (*S. l. var. cerasiforme*) in ten localities (Pajapan, Palenque, Coscomatepec, Xalapa, Tenejapan, Mahuixtlan, Tuxpan, Ixhuatlan, Maltrata, Ocotitlan, and Cereza) in the north, center and south of Veracruz, Mexico. The samples were labeled with the date of collection, place of collection (mountain, road, and village), geographic location (latitude, longitude and altitude) (Table 1). From each place where we collected, we obtained the values of the following variables: average annual temperature, diurnal average range, isothermality (Bio3), temperature seasonality (Bio4), annual precipitation (Bio12), precipitation of the driest months (Bio14), Seasonality of precipitation (Bio15), Precipitation of the warmest quarter (Bio18). The environmental variables was obtained of Worldclim project (Hijmans et al., 2005).

Morphological characterization in greenhouse

We conducted the investigation in a greenhouse located in the Faculty of Biological and Agricultural Sciences, located in Peñuela, Veracruz. We sow seeds from ten localities (plus a commercial variable as reference) in peat moss substrate in 80 cavity trays. Forty days after sowing, seedlings were transplanted in bags of size 32 x 30; We conducted the investigation under a completely randomized experimental design (Sanjuan-Lara et al., 2014) with four repetitions and eight plants each.

A nutritive N-P-K 20-20-20 formula was supplied, mixed with balanced microelements to promote plant development and daily growth. For the pest and disease prevention, products like Captan® and Confidor® were applied. For the morphological and agronomical characterization, 17 quantitative and 15 qualitative variables were evaluated according to the descriptors of IPGRI International Plant Genetic Resources Institute (IPGRI, 1996). For the measurement of the SD, ED and PD a digital standard and

Table 1: Coordinates, locations name and vegetative characterization of the locations

ID	Latitude	Longitude	Locality	Vegetation
1	20.951682	-97.460643	Tuxpan	Mangrove, Savannah and grassland with some dominant species of <i>Crescentia cujete</i> , <i>Curatella americana</i> , <i>Byrsonima crassifolia</i> , <i>Sabal mexicana</i> , <i>Quercus oleoides</i> ; agricultural activities: orange cultivation and pasture for livestock.
2	18.810362	-97.277332	Maltrata	Oak forest, high mountain coniferous and humid forests, species like: <i>Enterolobium cyclocarpo</i> , <i>Tabebuia pentaphylla</i> , <i>Holciocereus</i> sp. Crops like: Coffee, banana, lemon Orange and squash.
3	19.044236	-96.908142	Ocotitlán	
4	19.047061	-96.972935	Ixhuatlán del Café	
5	19.1644444	- 97.081666	Palenque	
6	19.413464	-96.915741	Mahuixtlán	
7	19.410983	-96.919018	Xalapa	
8	19.042595	-97.069957	Coscomatepec	
9	18.808572	-97.092217	Tenejapán	High evergreen forests, species such as <i>Bursera simaruba</i> , <i>Cupania dentata</i> , <i>Ficus</i> sp., <i>Dentropanax arboreus</i> . Crops like: Corn, bean, pineapple and pasture crops for livestock
10	18.2662183	-94.681884	Pajapán	

millimeter Vernier caliper was used. The PH, LL and LW traits were measured a model FH-3M flexometer (Table 2). We used a colorimeter CR 400 to identify the color of the fruits calculating the values of $L^*a^*b^*$, where L^* indicates the lightness, a^* represents the red/green coordinate and b^* is the yellow/blue coordinate (D'Souza et al., 1992).

Statistical analysis

We calculate the average of the quantitative variables and we carried out a principal component analysis (PCA) proposed by Pearson (1901), Esbensen and Geladi (1987); to determine which morphologic variables were the most important (Restrepo et al., 2012). Then we chose the variables with the greatest weight. After that we found 11 variables describing the variability of the collections (Fig. 1; Annex 1). A one-way anova as suggested by Wold, (1989) was performed for these 11 variables selected between all accessions coming from different localities. A post hoc analysis was followed a Tukey test ($p < 0.05$), we also determined the variation coefficients as suggested by Gomez and Gomez (1984). A cluster analysis (Broccard and Rudaz, 2012) was made by Ward's method to classify collections with high similarity into the same groups, we used the statistical package R (Li and Yan, 2018). A frequency table was made with the data of the qualitative variables to characterize the collections.

To determine the relation between the morphologic and environmental variables in the different localities of collections, we performed a descriptive analysis with the environmental variables (Tukey, 1977), and we also performed partial least square (PLS) analysis for vegetative, reproductive and yield variables (Geladi, 1988).

RESULTS

Regarding the PCA, the first two components explained 79 % variation, the first component contributed with 64.5 %

Table 2: Quantitative and qualitative variables evaluated in morphological characterization of wild populations of *Solanum lycopersicum* var. *cerasiforme*

Quantitative Variable	Qualitative variable
Plant height= PH	Terminal form of flowering= FTFF
Stem diameter= SD	Shoulder shape= FORH
Internode distance (cm) = ID	Fruit pubescence= PUBF
Number days to flowering= NDTFL	Pulp color= COLCA
Number of flowers per plant= NFP	Colour intensity= INTCOL
Number of clusters per plant	Heart color= COLCOR
Sepals length= SL	Cross section shape= FORCTF
Petals length= PL	Pistil scar shape= FORCPIS
Style length= StL	Terminal shape of fruit= FORTF
Staminal column length StmL	Firmness of the fruit= FIRMF
Pericarp thickness= PTh	Inflorescence type= TIPINFLO
Leaf area= LA	Pistil position= POSPIS
Leaf length= LL	Leaf position= POSH
Leaf width= LW	Fruit shape= FORFRUT
Number of leaflets= NLts	Growth type= TIPCREC
Fruit weight= FW	
Production/Plant=PP	

and the second with 14.1 % of the variation. The variables fruit weight, fruit size, leaf area, pericarp thickness, production per plant had the highest variation in the first component, while the variables stem diameter, number of flowers per plant, and sepal length and stamens length were the ones that contributed with the highest variation in the second component (Fig. 1, Annex 1). These were the most important morphologic variables according to the PCA.

Among the different localities, the analysis of variance indicated significant differences ($p < 0.05$) for all variables except for and flower number per plant; high coefficients of variation were recorded for the variables fruit weight, production per plant, number of flowers, length of style and leaf area (Table 3). The morphologic variables with

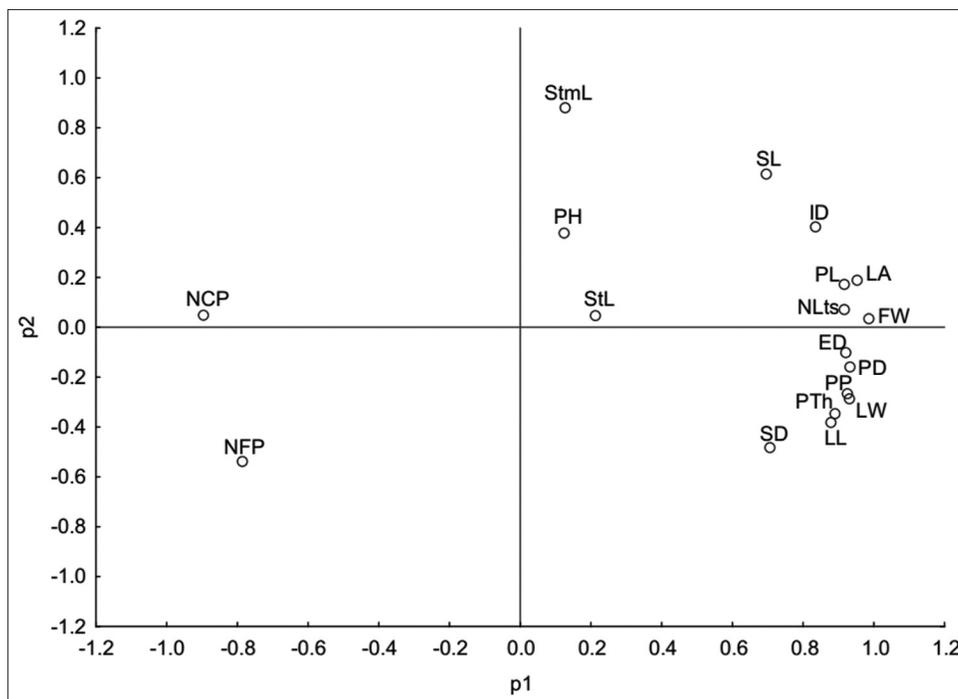


Fig 1. Quantitative variables from 11 collections of *S. l. var. cerasiforme* from Veracruz State based on the CPA (principal component analysis).

Table 3: Means comparison of quantitative variables of 11 collections of *S. l. var. cerasiforme*. Stem Diameter (mm) = SD, Internode distance (cm) = ID, Leaf area (cm²) = LA, Fruit weight = FW, Polar Diameter (mm) = PD, Means comparison of quantitative variables of 11 collections of *S. l. var. cerasiforme*. Equatorial Diameter (mm) = ED, Production/Plant = PP, Sepal's length = SL, Style Length (cm) = StL, Stamen's length (cm) = StmL, Number of flowers/plants = NFP, CV: coefficient of variation. Tukey (p < 0.05)

Collection	SD	ID	LA	FW	PD	ED	PP	SL	StmL	NFP	PTh
Pajapan	9.6 ^a	38.33 ^{bc}	179.4 ^a	9.54 ^a	32.64 ^a	33.93 ^a	983.7 ^a	0.67 ^{bc}	0.79 ^{ab}	30.14 ^{bc}	3.00 ^a
Cosco	9.1 ^{ab}	46 ^{ab}	160.7 ^{ab}	7.56 ^b	23.29 ^b	23.77 ^b	893.1 ^a	0.71 ^{bc}	0.83 ^a	33.2 ^{abc}	2.36 ^{ab}
Palenque	9.0 ^{ab}	31.71 ^{cd}	97.77 ^c	3.02 ^d	17.17 ^c	17.12 ^c	607.1 ^b	0.55 ^{bc}	0.64 ^c	50.0 ^a	1.69 ^{cde}
Ixhuatlán	8.4 ^{ab}	37.57 ^{cd}	120.7 ^{bc}	5.92 ^c	23.54 ^b	24.75 ^b	579.0 ^b	0.74 ^{ab}	0.74 ^{ab}	34.0 ^{abc}	1.96 ^{bcd}
Ocotitlán	8.3 ^{ab}	32.25 ^{cd}	103.9 ^c	2.77 ^d	17.05 ^c	16.96 ^c	636.9 ^b	0.52 ^c	0.68 ^{bc}	47.13 ^{ab}	2.06 ^{bc}
Mahuixtlan	8.4 ^{ab}	29.63 ^c	83.79 ^c	2.18 ^d	14.45 ^c	15.83 ^c	192.6 ^c	0.61 ^{bc}	0.81 ^a	36.3 ^{abc}	1.28 ^e
Tenejapan	8.2 ^{ab}	47.86 ^a	179.4 ^a	7.66 ^b	22.31 ^b	23.44 ^b	886.0 ^a	0.94 ^a	0.87 ^a	20.43 ^c	2.00 ^{bc}
Tuxpan	8.1 ^{ab}	29.63 ^c	95.42 ^c	1.825 ^c	14.70 ^c	15.55 ^c	180.0 ^c	0.63 ^{bc}	0.84 ^a	42.13 ^{ab}	1.25 ^e
Xalapa	8.1 ^{ab}	31 ^{cd}	94.01 ^c	2.57 ^d	15.10 ^c	16.04 ^c	200.1 ^c	0.61 ^{bc}	0.81 ^a	44.13 ^{ab}	1.29 ^{de}
Maltrata	7.7 ^b	31.25 ^{cd}	90.98 ^c	1.946 ^c	15.27 ^c	16.31 ^c	301.5 ^c	0.55 ^{bc}	0.81 ^a	47.63 ^a	1.30 ^{de}
Cereza	7.5 ^b	35.71 ^{cd}	96.35 ^c	1.95 ^c	14.37 ^c	16.14 ^c	195.3 ^c	0.60 ^{bc}	0.79 ^{ab}	43.14 ^{ab}	1.00 ^e
CV	12.6	21.23	35.36	67.09	29.75	30.69	62.09	24.17	12.18	34.02	36.84

the highest coefficient of variation between the collections were LA, FW, ED, PD, PP and NFP respectively 35.3 %, 67.9 %, 29.7 %, 30.6 %, 62 % and 34 % respectively. By contrast those with the smallest variation were SD, and StmL respectively 12.6 % and 12.1 % (Table 3). Collections from Pajapan, Coscomatepec, Ixhuatlán del Café and Tenejapan showed greater fruit size, production per plant, leaf area development but fewer flowers per plant. The mean weight of fruit registered was high in the collections of Pajapan (8.87 g) followed by Coscomatepec (7.17 g); the two collections that registered lower average of fruit weight were Tuxpan with 1.82 g and Maltrata with 1.94 g. The collection coming from Ixhuatlán del Café is the only

one that presented flowers with an exert style which would result in interspecific crossing. In the collections we found that 91 % had the style inserted and 9 % had an exert style flower (Table 4). As for the qualitative variables, we found that 45 % of the collections presented indeterminate type growth, the other determined growth (Table 4). The shape of the pistil scar was dotted for 82 % of the collections and stellated for 18 % (Table 4). We registered three leaf positions in the plant (inclined, semi-erect and horizontal), 82 % presented inclined leaf, 9 % semi-erect and 9 % horizontal (Table 4). The corolla was yellow in 82 %, orange in 18 %. Regarding the color of the fruits 46 % presented a red color with a high intensity, 36 %

Table 4: Presentation of the qualitative variables based on categorical variables from IPGRI indicators in the characterization of 11 collections of *S. l. var. cerasiforme*. Terminal form of flowering = FTFF, Shoulder shape = FORH, Fruit pubescence = PUBF, Fruit pubescence = PUBF, Pulp color = COLCA, Color intensity = INTCOL, Heart color = COLCOR, Cross section shape = FORCTF, Pistil scar shape = FORCPIS, Terminal shape of fruit = FORTF, Presentation of the qualitative variables based on categorical variables from IPGRI indicators in the characterization of 11 collections of *S. l. var. cerasiforme*. Firmness of the fruit = FIRMF, Inflorescence type = TIPINFLO, Pistil position = POSPIS, Leaf position = POSH, Fruit shape = FORFRUT, Growth type = TIPCREC

Collection	FTFF	FORH	PUBF	COLCA	INTCOL	COLCOR	FORCTF	FORCPIS	FORTF	FIRMF	POSH	TIPINFLO	FORFRUT	POSPIS	TIPCREC
Pajapan	2	3	3	5	7	1	1	1	2	7	7	3	1	1	2
Cosco	2	3	3	5	7	1	1	1	2	7	5	3	3	1	2
Palenque	2	3	3	5	5	1	1	2	2	7	7	3	3	1	4
Ixhuatlan	2	3	3	5	5	1	1	2	2	7	7	3	3	4	4
Ocotitlan	2	3	3	5	5	1	1	1	2	7	3	3	3	1	2
Mahuixtlan	2	3	3	5	3	1	1	1	2	7	7	3	3	1	4
Tenejapan	2	3	3	5	5	1	1	1	2	7	7	3	1	1	2
Tuxpan	2	3	3	5	7	1	1	1	2	7	7	3	1	1	4
Xalapa	2	3	3	5	3	3	1	1	2	7	7	3	3	1	4
Maltrata	2	3	3	5	7	1	1	1	2	7	7	3	3	1	4
Cereza	2	3	3	5	7	5	1	1	2	7	7	3	3	1	2

of red fruits with moderate intensity and 18% with low intensity (Table 4). In this investigation regarding the fruit shape, we found two types: 73 % round and 27 % flattened (Table 4), which coincided with the report by Fernandes et al. (2018). Table 3. Means comparison of quantitative variables of 11 collections of *S. l. var. cerasiforme*. Stem Diameter (mm) = SD, Internode distance (cm) = ID, Leaf area (cm²) = LA, Fruit weight = FW, Polar Diameter (mm) = PD, Means comparison of quantitative variables of 11 collections of *S. l. var. cerasiforme*. Equatorial Diameter (mm) = ED, Production/Plant = PP, Sepal's length = SL, Style Length (cm) = StL, Stamen's length (cm) = StmL, Number of flowers/plants = NFP, CV: coefficient of variation. Tukey (p < 0.05).

In the south region of the state, we collected the species mean annual temperature between 22.7 to 24.8, isothermality ranged from 55 to 56 and temperature seasonality from 2099 to 2290. The precipitation, precipitation seasonality ranged respectively from 2041 to 2357 mm, from 69 to 73. The soil of the south region was characterized by concentrations of K from 476 to 526 cmol/L, Mg from 0.033 to 0.037, Na absorption 3.47 to 11.68, pH 5.9 to 6.92. The center was characterized by a mean annual temperature from 15 to 23, isothermality from 59 to 69 and temperature seasonality from 1738 to 2186 while we registered a precipitation from 824 to 2228 mm and precipitation seasonality from 67 to 83. The concentrations of K, Mg ranged from 0.036 to 320 cmol/L, 0.124 to 0.970 cmol/L respectively. The North region was characterized by a mean annual temperature from 24.2 to 24.4, isothermality 51 and temperature seasonality from 3247 to 3295. The precipitation ranged from 1151 to 1179 mm, precipitation seasonality from 36 to 65. In the north region of the state the concentration of K, Mg, ranged from 287 to 305 cmol/L and 0.033 to 0.079 cmol/L; the pH ranged from 6 to 7.02 and the Na absorption from 3.48 to 4.46 (Table 5, 6).

The analysis of clusters detected four groups, the first group contained by three collections Tenejapan, Pajapan, and Coscomatepec. The second group contained five collections Xalapa, Mahuixtlan, Tuxpan, Maltrata, and Cherry. Palenque and Ocotitlan formed the third group. The fourth group is formed by a single collection Ixhuatlan del café. The CPA showed the same clustering pattern as cluster analysis (Fig. 2 A, B).

Based on the PLS analysis, we observed that the vegetative variables (SD, LA and ID) were related positively to Bio1, Bio4, Bio12 for the localities like Ocotitlan, Ixhuatlan, Cosco, Tenejapan. We observed lower influence of those environmental variables in Maltrata, Mahuixtlan, Tuxpan (Fig. 3 A). While we found great correlation between Bio1, Bio12 and K with the productive variables (PP, FW)

Table 5: Environmental characterization of the northern, central and southern regions of Veracruz, Altitude (m.a.s.l), Bio1 = mean annual temperature (°C), Bio2 = mean diurnal range (°C), Bio3 = isothermality, Bio4 = temperature seasonality, Bio12 = annual precipitation (mm), Bio14 = precipitation of the driest month (mm), Bio15 = precipitation seasonality (mm), Bio18 = precipitation of the warmest quarter, mm)

Variable	Altitude	Bio1	Bio2	Bio3	Bio4	Bio12	Bio14	Bio15	Bio18	Region
Mean	18.5	24.3	9.1	51.0	3265.0	1166.5	28.0	64.0	98.3	North
Minimum	14.0	24.2	9.0	51.0	3247.0	1151.0	27.0	63.0	96.0	
Maximum	24.0	24.4	9.0	51.0	3295.0	1179.0	30.0	65.0	101.0	
SD	4.4	0.08	0.12	0	20.7	12.0	1.4	0.8	2.1	
Mean	1346.5	18.8	11.8	64.1	1953.4	1791.4	41.5	76.0	130.8	Center
Minimum	553.0	15.1	10.0	59.0	1738.0	824.0	12.0	67.0	41.0	
Maximum	2100	23	13.9	69.0	2186.0	2228.0	55.0	83.0	169.0	
SD	462.0	2.1	1.1	2.93	145.4	353.0	11.7	3.9	34.7	
Mean	392.3	23.6	9.2	55.5	2176.5	2260.5	43.0	70.0	155.0	South
Minimum	138	22.7	8.9	55.0	2099.0	2041.0	39.0	69.0	127.0	
Maximum	541.0	24.8	9.6	56.0	2290.0	2357.0	46.0	73.0	176.0	
SD	192.152	1.034	0.34	0.577	94.193	147.261	3.559	2.0	24.9	

Table 6: Nutritional characterization of soils (North, center, and South) region of Veracruz

Variable	K (cmol / L)	Mg (cmol / L)	MO	Na (absorption ratio)	pH	Region
Mean	293.750	0.055	0.203	3.941	6.51	North
Minimum	287.000	0.033	0.189	3.482	6	
Maximum	305.000	0.079	0.216	4.462	7.02	
DS	7.890	0.019	0.011	0.403	0.49	Center
Mean	0.110	0.311	4.647	0.238	5.921	
Minimum	0.036	0.124	3.057	0.076	5.020	
Maximum	0.320	0.970	7.530	0.933	7.017	
DS	0.088	0.214	1.494	0.181	0.555	South
Mean	508.500	0.032	0.410	5.947	6.41	
Minimum	476.000	0.030	0.371	3.478	5.9	
Maximum	526.000	0.037	0.494	11.688	6.92	
DS	23.629	0.003	0.057	3.847	0.49	

(Fig.3 A) in Ixhuatlan and Ocotitlan. The environmental variables Bio3, Bio2, Bio15 showed high correlation with variables SL, NFP in Tuxpan, Mahuixtlan while StI showed strong variation with Bio14 in Pajapan (Fig. 3 B).

DISCUSSION

The fact that the collections differ one to another in one or more variables, implies a phenotypic divergence between the collections. According to fruit length our results are close to those found by Carrillo Rodríguez and Chávez Servia (2010), who reported fruit length of 2.6 and 3.1 cm, respectively.

Regarding equatorial fruit diameter, similar patterns were also observed in several other studies conducted by Álvarez-Hernández et al. (2009) and Carrillo Rodríguez and Chávez Servia (2010), with respective equatorial diameters from 2.1 to 2.4 cm in the larger fruits, and 1 to 1.1 cm in the smaller ones, and from 2.8 to 3.7 cm in the larger fruits, less than 1.8 cm in the smaller ones. In terms of fruit size,

our results were lower than those reported by the authors previously mentioned. This could have been due to the intrinsic characteristics of the collections. The tomato red color indicates the presence of lycopene, which is the most abundant carotenoid in the fruit (Saini et al., 2015), this compound according to Leong et al. (2018) is very important in the prevention of cancer and cardiovascular diseases. Vargas et al. (2015), in their genetic characterization of various tomato accessions, reported intense yellow, pink and red fruits. (Agudelo and Aguirre, 2011) in the characterization of cherry tomatoes reported three groups of fruits according to the color (red, yellow, pink). The shape and size of the tomato fruit are properties of great concern both for the consumer and for the transportation; shape is an attribute that allows the visualization at a single glance and accurate identification of tomato cultivars in the field inspections. However, Rocha et al. (2010) reported five shapes of fruit piriform, cylindrical, cordiform, slightly flattened, and globular. Agudelo et al. 2011 found 37 % round, 26 % flattened, 21 % cylindrical and 16 % elongated. We found low fruit variation in relation to the information reported by (Bhattarai et al., 2018) and Salim et al. (2018), who respectively found flattened, slightly flattened, cylindrical, rounded, high rounded and rounded, flattened, ellipsoidal and heart-shaped fruits. This was because they carried out morphological characterization between different tomato species, whereas in this study the characterization between populations within a species was carried out. It is interesting to mention that fruit shape is a stable trait that is not influenced by biotic and abiotic stress, so it is an important character or trait that allow clear differentiation in cultivars of tomato (García-Gusano et al., 2004; Vishwanath et al., 2014) and in many other tomato species (Arya and Saini, 1976; Patel et al., 2001).

The position of the style is a very important character in the determination of the type and time of pollination of

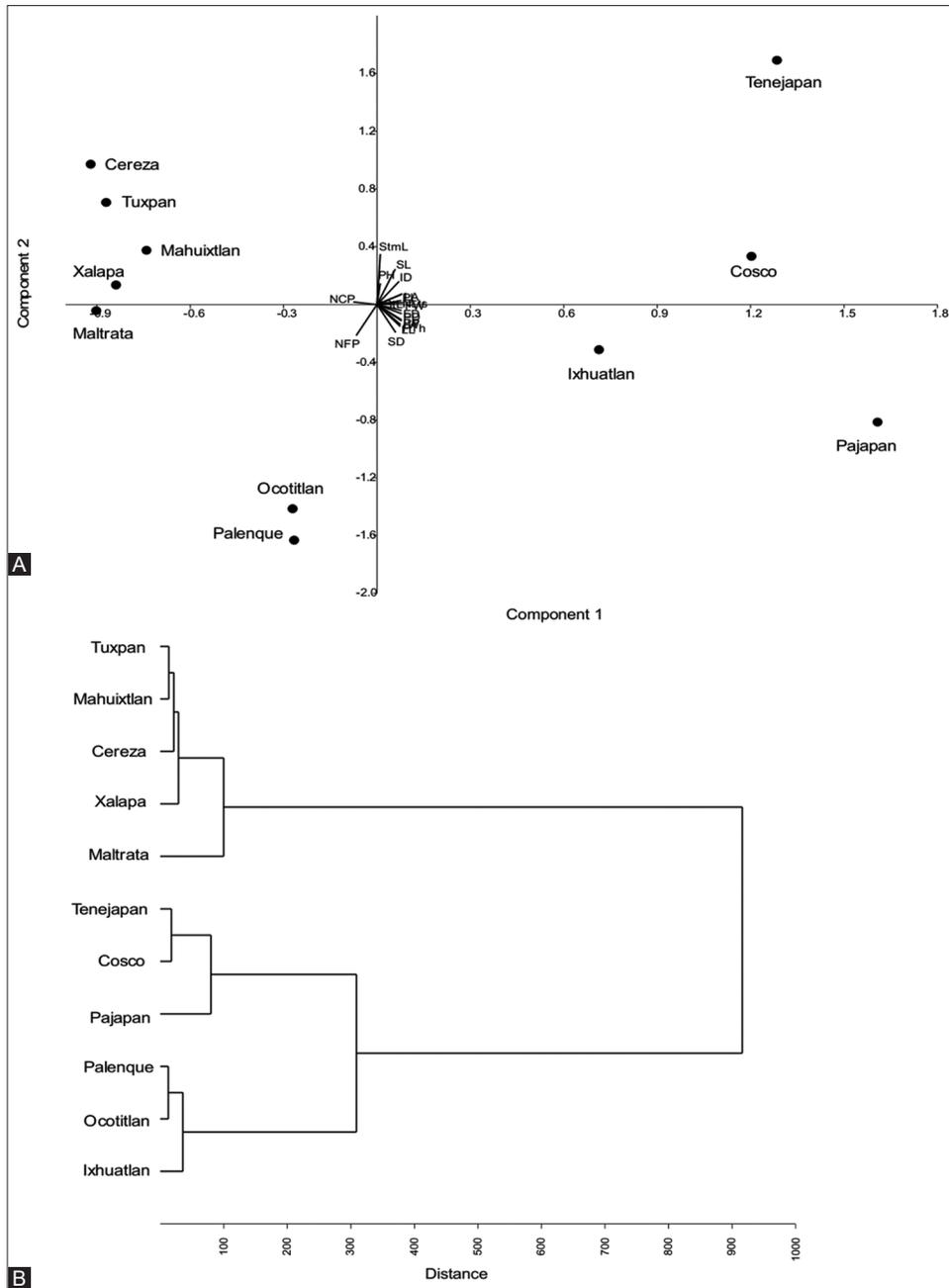


Fig 2. Grouping of collections according to CPA and cluster analysis based on their similarity.

the plant; it was very determinant in characterizing the collections of Ixhuatlan del Cafe in this work. Mata-Nicolás et al., 2020 already reported that *S. l. var. cerasiforme* from Mexico presented diversity regarding the position of the style. The results reported in this study, matched with Yesmin et al. (2014) and Vishwanath et al. (2014) who also reported two types of style position on the flower. We registered variation in the length of the stamen in the collections; it is an important character in the identification of varieties.

It is necessary to mention that Yesmin et al. (2014) reported two leaf positions within 11 accessions while Salim et al.

(2018) found three positions (59.09 % horizontal, 27.27 % semi-right and 13.64 % inclined). The collections that we studied showed greater variation in leaf position variable than those reported by Yesmin et al. (2014). The plant height is an important component of plant architecture, and is strongly correlated with the yield, it is affected by the internode length (Salas Fernandez et al., 2009). However, we didn't register important variation regarding plant height, but we found greater variation between collections in internode length variable, it is a key variable when we know that there is strong relation with harvest index and biomass production according to Sun et al.

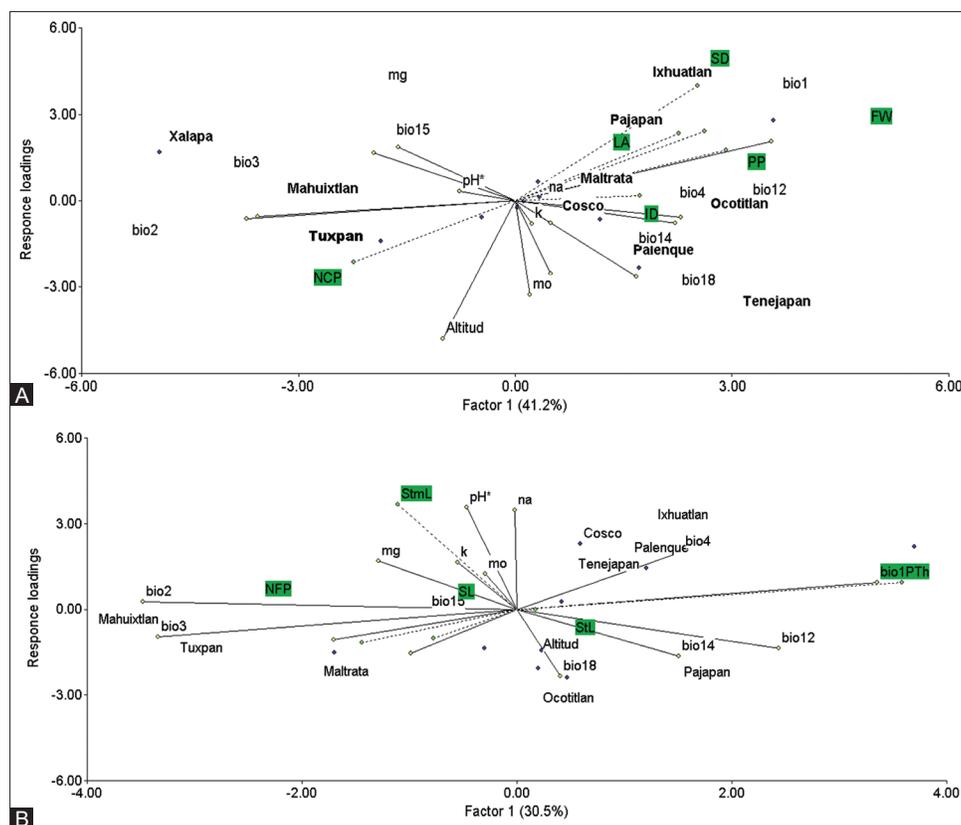


Fig 3. determination of interaction between phenotypic variables A (SD, LA, FW, PP, NCP, ID); B (StmI, STI, SL, NFP, PTh) and environmental variables.

(2019). Internode is one of the factors affecting the plant height (Sun et al., 2019); it is very sensitive to the effects of environmental stresses like high temperature, lack sunlight and excess of nitrogen (Yamamoto et al., 2016).

Bonilla-Barrientos et al. (2014) found important variation in yield and reproductive variables when they conducted the morphological characterization of pepper tomatoes and kidney-shaped tomatoes, they reported that fruit weight registered the highest variation. Restrepo et al. (2006), in wild germplasm of *Lycopersicon* spp. reported that the variables of reproduction (flowers) and vegetative development (stem diameter) explained 76 % of the total variation in the first two components. It is important to mention that a commercial variety has been used, afterwards we conducted de experiment with 11collections. When considering the results found in this work, the morphologic variation between the collections and those reported by Bonilla-Barrientos et al. (2014) who evaluated morphologic characterization in native tomatoes, Moya et al. (2005) which studied morphologic variability among varieties of cultivated tomato we observed the same pattern of variability. From our collections we detected a large of diversity for evaluated traits like leaves, fruit weight and number of flowers. We found high variations as our collections were found in a wide geographic and environmental range.

When comparing the climatic variables of different regions of collections in this experiment, we found different range in Bio1, Bio4, Bio12, Bio15. This climatic differentiation could lead to phenotypic variability because of the high plasticity of the species that we studied. The north region presents less precipitation, but it showed more stability than the center and south region which showed more precipitation more variability. In other words, the precipitation varied too much during the year. Regarding the variables of temperature, we found similar mean annual temperature in the south and north region of the state. We observed greater range of altitude in the center region, the elevation is a variable that can take influence in all the other temperature and precipitation variables. In this part of the state, we collected plant a different altitude. Regarding the environmental variables of temperature, Vargas et al. (2005) recorded tomato plants at an average annual temperature between 18 ° C and 22 ° C in the domestication area, while Nakazato et al. (2008) reported an average annual temperature of 20.8 ° C in South America. In the case of precipitation variables, Vargas et al. (2005) reported precipitation between 800 and 1 000 mm; Álvarez-Hernández et al. (2009) reported from 751 to 1014 mm; and in the present work we registered the species with precipitation 1166.5, 1791.4 and 2176.5 mm respectively

in north, center, south region of the state All these values are lower than those reported for the center of origin such as that of Nakazato et al. (2010) of 1800 mm and those found in (1768 mm).

CONCLUSION

The morphologic traits showed great variation between the localities, they evidenced phenotypic diversity. The area of collections of the species presented different climatic conditions. This study showed intraspecific morphological differences between wild tomato populations *S. l.* var. *cerasiforme*, in the state of Veracruz, a state with a wide environmental and ecosystem diversity. The variables that produced the greatest weight in the phenotypic variation between collections were fruit weight, fruit size, pericarp thickness, leaf area and number of leaflets. We found great correlations between the environmental and morphologic variables, but we did not figure out if these correlations are causal. The collections we have studied could constitute a valuable germplasm bank for genetic improvement programs.

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REFERENCES

Agong, S. G., S. Schittenhelm and W. Friedt. 2001. Genotypic variation of Kenyan tomato (*Lycopersicon esculentum* L.) germplasm. J. Food Technol. Afr. 6: 13-17.

Agudelo, A. G. A. and N. C. Aguirre. 2011. Caracterización morfológica del tomate tipo cereza (*Solanum lycopersicum* Linnaeus). -53 Available from: <http://www.vip.ucaldas.edu.co/agronomia/>

downloads.

Álvarez-Hernández, J. C., H. Cortez-Madrigal and I. García-Ruiz. 2009. Exploración y caracterización de poblaciones silvestres de jitomate (*Solanaceae*) en tres regiones de michoacán, México. Polibotánica. 28: 139-159.

Arya, P. S. and S. S. Saini. 1976. Genetic variability and correlation studies in bell peppers. Haryana J. Hortic. Sci. 5: 236-244.

Bai, Y. and P. Lindhout. 2007. Domestication and breeding of tomatoes: What have we gained and what can we gain in the future? Ann. Bot. 100: 1085-1094.

Bauchet, G. and M. Causse. 2012. In: ,M., Genetic Diversity in Tomato (*Solanum lycopersicum*) and its Wild Relatives, Genetic Diversity in Plants. In Tech, London. Available from: <http://www.intechopen.com/books/genetic-diversity-in-plants/genetic-diversity-in-tomatosolanum-lycopersicum-and-its-wild-relatives>.

Bergougnoux, V. 2014. The history of tomato: From domestication to bio-pharming. Biotechnol. Adv. 32: 170-189.

Bhattarai, K., S. Sharma and D. R. Panthee. 2018. Diversity among modern tomato genotypes at different levels in fresh-market breeding. Int. J. Agron. 2018: 1-15.

Blanca, J., J. Cañizares, L. Cordero, L. Pascual, M. J. Diez and F. Nuez. 2012. Variation revealed by SNP genotyping and morphology provides insight into the origin of the tomato. PLoS One. 7: e48198.

Bonilla-Barrientos, O., R. Lobato-Ortiz, J. J. García-Zavala, S. Cruz-Izquierdo, D. Reyes-López and E. Hernández-Leal. 2014. Diversidad agronómica y morfológica de tomates arriñonados y tipo pimienta de uso local en Puebla y Oaxaca, México. Rev. Fitotec. Mex. 37: 129.

Carrillo-Rodríguez, J. C. and J. L. Chávez-Servia. 2010. Caracterización agromorfológica de muestras de tomate de Oaxaca. Rev. Fitotec. Mex. 33: 1-6.

Crisanto-Juárez, A. U., A. M. Vera-Guzmán, J. L. Chávez-Servia and J. C. Carrillo-Rodríguez. 2010. Calidad de frutos de tomates silvestres (*Lycopersicon esculentum* var. *cerasiforme* Dunal) de Oaxaca, México. Rev. Fitotec. Mex. 33: 7-13.

Délices, G., R. Otto, R. N. Pastrana, P. A. Meza, R. Serna-Lagunez and R. G. Pastrana. 2019. Biogeografía del tomate *Solanum lycopersicum* var. *cerasiforme* (*Solanaceae*) en su centro de origen (sur de América) y de domesticación (México). Rev. Biol. Trop. 67: 1023-1036.

Dharmatti, P. R., B. B. Madalgeri, I. M. Mannikeri, R. V. Patil and G. Patil. 2001. Genetic divergence studies in summer tomatoes. Karnataka J. Agric. Sci. 14: 407-411.

D'Souza, M. C., S. Singha and M. Ingle. 1992. Lycopene concentration of tomato fruit can be estimated from chromaticity values. HortScience. 27: 465-466.

Fernandes, M.O., P. A. Bianchi, L. R. A. Silva, L. S. Vianna, Santos and Moulin. 2018. Morpho-agronomic characterization and analysis of genetic divergence among accessions of tomatoes (*Solanum lycopersicum* L.). Cienc. Rural. 48.

Fischer, I., L. Camus-Kulandaivelu, F. Allal and W. Stephan. 2011. Adaptation to drought in two wild tomato species: The evolution of the ASR gene family. New Phytol. 190: 1032-1044.

Flores-Hernández, L. A., R. Lobato-Ortiz, J. J. García-Zavala, J. D. Molina-Galán, D. M. Sargerman-Jarquín and M. D. J. Velasco-Alvarado. 2017. Parientes silvestres del tomate como fuente de germoplasma para el mejoramiento genético de la especie. Rev. Fitotec. Mex. 40: 83-91.

Foolad, M. R. 2007. Current status of breeding tomatoes for salt and drought tolerance. In: Jenks, M., P. Hasegawa and S. Jain, (Eds.), Advances in Molecular Breeding Toward Drought and

- Salt Tolerant Crops. Springer, Berlin, pp. 669-700.
- García-Gusano, M., S. García-Martínez and J. J. Ruiz. 2004. Use of SNP markers to genotype commercial hybrids and Spanish local cultivars of tomato. *Tomato Genet. Coop Rep.* 54: 12-15.
- García-Martínez, S., L. Andreani, M. Garcia-Gusano, F. Geuna and J. J. Ruiz. 2006. Evaluation of amplified fragment length polymorphism and simple sequence repeats for tomato germplasm fingerprinting: Utility for grouping closely related traditional cultivars. *Genome.* 49: 648-656.
- Geladi, P. 1988. Notes on the history and nature of partial least squares (PLS) modelling. *J. Chemometr.* 2: 231-246.
- Gomez, K. A. and A. A. Gomez. 1984. *Statistical Procedures for Agricultural Research.* International Rice Research Institute, College. John Wiley & Sons, New York.
- González-Aguilera, J., L. A. Pessoni, G. Belfort-Rodrigues, A. Y. Elsayed, D. J. Henriques-da Silva and E. Gonçalves-de Barros. 2011. Genetic variability by ISSR markers in tomato (*Solanum lycopersicum* Mill.). *Rev. Bras. Cienc. Agrarias.* 6: 243-252.
- Hijmans, R. J., S. E. Cameron, J. L. Parra, P. G. Jones and A. Jarvis. 2005. Very high-resolution interpolated climate surfaces for global land areas. *Int. J. Climatol.* 25: 1965-1978.
- Jenkins, J. A. 1948. The origin of the cultivated tomato. *Econ. Bot.* 2: 379-392.
- Leong, H. Y., P. L. Show, M. H. Lim, C. W. Ooi and T. C. Ling. 2018. Natural red pigments from plants and their health benefits: A review. *Food Rev. Int.* 34: 463-482.
- Li, K. and E. Yan. 2018. Co-mention network of R packages: Scientific impact and clustering structure. *J. Informetr.* 12: 87-100.
- Liu, D., L. Yang, J. Zhang, G. Zhu, H. Lü and Y. Lü. 2020. Domestication and breeding changed tomato fruit transcriptome. *J. Integr. Agric.* 19: 120-132.
- Mata-Nicolás, E., J. Montero-Pau, E. GimenoPaez, V. Garcia-Carpintero, P. Ziarolo and N. Menda. 2020. Exploiting the diversity of tomato: The development of a phenotypically and genetically detailed germplasm collection. *Hortic. Res.* 7: 1-14.
- Mohanty, A., J. P. Martín and I. Aguinagalde. 2001. A population genetic analysis of chloroplast DNA in wild populations of *Prunus avium* L. in Europe. *Heredity.* 87: 421-427.
- Nakazato, T., D. L. Warren and L. C. Moyle. 2010. Ecological and geographic modes of species divergence in wild tomatoes. *Am. J. Bot.* 97: 680-693.
- Parthasarathy, V. A. and C. Aswath. 2002. Genetic diversity among tomato genotypes. *Indian J. Hortic.* 59: 162-166.
- Patel, D. A., P. T. Shukla and G. C. Jadeja. 2001. Morphological studies on interspecific hybrids between *Solanum indicum* L. and *Solanum melongena* L. *Indian J. Genet.* 61: 180-182.
- Pearson, K. 1901. On lines and planes of closest fit to systems of points in space. *Philos. Mag.* 2: 559-572.
- Peralta, I. E., S. Knapp and D. M. Spooner. 2005. New species of wild tomatoes (*Solanum* Section *Lycopersicon*: *Solanaceae*) from Northern Peru. *Syst. Bot.* 30: 424-434.
- Peralta, I. E., D. M. Spooner and S. Knapp. 2008. Taxonomy of wild tomatoes and their relatives (*Solanum* sect. *Lycopersicoides*, sect. *Juglandifolia*, sect. *Lycopersicon*; *Solanaceae*). *Syst. Bot.* 84: 186.
- Ranc, N., S. Muños, S. Santoni and M. Causse. 2008. A clarified position for *solanum lycopersicum* var. *cerasiforme* in the evolutionary history of tomatoes (*Solanaceae*). *BMC Plant Biol.* 8: 130.
- Restrepo, E. F. S., F. A. C. Vallejo and M. A. Lobo. 2006. Evaluación de la resistencia al pasador del fruto *Neoleucinodes elegantalis* y caracterización morfoagronómica de germoplasma silvestre de *Lycopersicon* spp. *Acta Agron.* 55: 15-21.
- Restrepo, L. F., L. S. Posada and R. Noguera. 2012. Application of the principal-component analysis in the evaluation of three grass varieties. *Rev. Colomb. Cienc. Pecuarias.* 25: 258-266.
- Rick, C. M. 1974. The tomato. In: King, R.C., (Ed.), *Handbook of Genetics: Plants, Plant Viruses, and Protists.* Springer, Boston, MA, pp. 247-280.
- Saavedra, T. M., G. A. Figueroa, J. G. D. Cauih, T. M. Saavedra, G. A. Figueroa and J. G. D. Cauih. 2017. Origin and evolution of tomato production *Lycopersicon esculentum* in México. *Ciênc. Rural.* 47: 3.
- Saini, R. K., S. H. Nile and S. W. Park. 2015. Carotenoids from fruits and vegetables: Chemistry, analysis, occurrence, bioavailability and biological activities. *Food Res. Int.* 76: 735-750.
- Salas Fernandez, M. G., P. W. Becraft, Y. Yin and T. Lübberstedt. 2009. From dwarves to giants? Plant height manipulation for biomass yield. *Trends Plant Sci.* 14: 454-461.
- Salim, M. M. R., M. H. Rashid, M. M. Hossain and M. Zakaria. 2018. Morphological characterization of tomato (*Solanum lycopersicum* L.) genotypes. *J. Saudi Soc. Agric. Sci.* 19: 233-240.
- Sanjuan-Lara, F., P. Ramírez-Vallejo, P. Sánchez-García, M. Livera-Muñoz, M. Sandoval-Villa, J. C. Carrillo-Rodríguez and C. Perales-Segovia. 2014. Variación en características de interés agronómico dentro de una población nativa de tomate (*Solanum lycopersicum* L.). *Rev. Fitotec. Mex.* 37: 159.
- Scintu, A., M. Rodriguez, D. Rau, J. Giovannoni and G. Attene. 2015. Characterization of a wide collection of tomato (*Solanum lycopersicum* L.) for morpho-phenological, quality and resistance traits. *J. Agric.* 21: 38-43.
- Stoilova, T. and G. Pereira. 2013. Assessment of the genetic diversity in a germplasm collection of cowpea (*Vigna unguiculata* (L.) Walp.) using morphological traits. *Afr. J. Agric. Res.* 8: 208-215.
- Sun, X., J. Shu, A. M. Ali Mohamed, X. Deng, X. Zhi, J. Bai, Y. Cui, X. Lu, Y. Du, X. Wang, Z. Huang, Y. Guo, L. Liu and J. Li. 2019. Identification and characterization of El (elongated internode) gene in tomato (*Solanum lycopersicum*). *Int. J. Mol. Sci.* 20: 2204.
- Vargas, T. O., E. P. Alves, A. C. Abboud, M. A. Leal and M. G. Carmo. 2015. Diversidade genética em acessos de tomateiro heirloom. *Hortic. Bras.* 33: 174-180.
- Vishwanath, K., P. S. Rajendra, H. M. Pallavi and K. P. R. Prasanna. 2014. Characterization of tomato cultivars based on morphological traits. *Ann. Plant Sci.* 3: 854-862.
- Wold, S., K. Esbensen and P. Geladi. 1987. Principal component analysis. *Chemometr. Intell. Lab. Syst.* 2: 37-52.
- Yamamoto, K., W. Guo and S. Ninomiya. 2016. Node detection and internode length estimation of tomato seedlings based on image analysis and machine learning. *Sensors (Basel).* 16: 1044.

ANNEX

Annex 1: Variable weight in the principal component analysis for 11 collections of *S. l. var. cerasiforme*. Only the first two components are listed

Variable	PC1	PC2
Stem diameter	0.209	0.375
Fruit weight	0.298	0.002
Polar diameter	0.281	0.130
Equatorial diameter	0.278	0.093
Plant height	0.033	-0.234
Production per plant	0.275	0.183
Distance to first inflorescence	0.255	-0.251
Number of flowers	-0.242	0.322
Number of clusters	-0.269	-0.056
Sepals length	0.214	-0.378
Petals length	0.275	-0.090
Style length	0.063	-0.006
Staminal length	0.049	-0.567
Leaf area	0.290	-0.106
Leaf length	0.257	-0.244
Leaf width	0.277	0.207
Number of leaflets	0.278	-0.029
Pericarp thickness	0.270	0.219
% variation	64.5	14.1
% accumulated variation		78.6



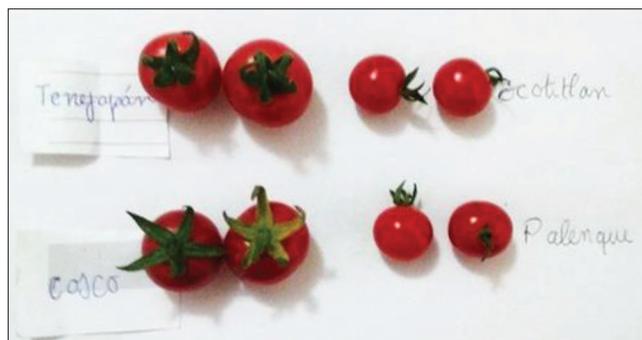
Annex 3. Picture of tomatoes fruit collected during the field work.



Annex 4. Picture of tomato flowers collected during the field work.



Annex 2. Pictures during the field work



Annex 5. Picture of tomatoes fruit collected under greenhouse conditions.