Australian Journal of Crop Science

AJCS 7(7):933-940 (2013)

AJCS ISSN:1835-2707

Yield and fruit quality evaluation in husk tomato autotetraploids (*Physalis ixocarpa*) and diploids

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Abstract

Tomatillo (*Physalis ixocarpa* Brot.) is a native species of Mexico and Central America, ranking fourth in cultivated area among main vegetables in Mexico. The objective was to evaluate and select, husk tomato autotetraploid and diploid populations for yield and fruit quality. Five autotetraploid populations with higher yields and fruit quality, previously selected for an assessment study of autotetraploids formed by the action of colchicines were used. The five autotetraploid populations were compared with four diploid populations with outstanding agronomic traits in the source regions. The populations were established under a completely randomized block design with four replications. The studied traits were: Fruit yield, fruit weight, fruits per plant, equatorial and polar fruit diameter, total soluble fruit solids, fruit firmness, pH and vitamin C content. The diploids showed the following characteristics; fruit yield=1.809 kg plant⁻¹; number of fruits per plant=56.227. The autotetraploid presented fruit yield = 1.688 kg plant⁻¹, number of fruits per plant = 60.776. Analysis of variance showed significant differences between locations, and between populations for most variables, indicating that they responded in a different way to the environments and to both ploidy levels under study. When comparing the group of four diploid populations with the five autotetraploids, significant differences ($p \le 0.01$) were found for polar and equatorial diameter of fruit, and fruit firmness ($p \le 0.05$), for vitamin C, however when comparing the Rendidora diploid with the group of five autotetraploids, significant differences were found ($p \le 0.01$) for fruit weight, fruit equatorial and polar diameters, fruit firmness and vitamin C content, and ($p \le 0.05$) for fruit yield per plant and number of fruits per plant. The chromosomal duplication in husk tomato Physalis ixocarpa contributed to the change in the shape of the fruits which flattened at the poles with the presence of gaps between the endocarp and the mesocarp. The vitamin C content in autotetraploids was enhanced which contributed to the improvement of the fruit nutritional quality. It is noteworthy that the evaluated autotetraploids are materials needing a process of selection and chromosomal stabilization in order to achieve the full expression of their genetic potential.

Keywords: *Physalis ixocarpa*, autopoliploidy, genetic variability, fruit yield, plant breeding. **Abbreviations:** AFW_average fruit weight, EFD_equatorial fruit diameter, FF_fruit firmness, FY_fruit yield, NFP_Number of fruits per plant, PFD_polar fruit diameter, TSS_total soluble solids, VC_vitamin C content.

Introduction

Husk tomato (*Physalis ixocarpa* Brot. 2n = 2x= 24) is a species native to Mexico and Central America and it is, for the time being, one of the most important vegetable crops in Mexico (Cantwell et al., 1992) ranking in the fourth place in planted area (47.472 ha) among commercially cultivated vegetables (SIAP-SAGARPA, 2011). Its current importance is due to the significant increase in the *per capita* consumption (4.5 kg) at national level as well as to the increasing exports to the United States and Canada (Peña et al., 2002). Besides Mexico, husk tomato is grown in Guatemala, Colombia (Bukasov, 1963), Poland, Russia (Bock et al., 1995), Southeast United States, Israel, and South Africa (Peña and Márquez, 1990), the Rajasthan region in

India, Australia, Kenya, Africa, Bahamas, Puerto Rico, Jamaica, England and Taiwan (Morton, 1987).The most important countries, after Mexico, in the cultivation of this species are New Zealand, Australia, Africa, Kenya and India (Fisher et al., 1990). Although there is wide genetic variability in the husk tomato (Santiaguillo et al., 2004), the genetic improvement of tomato species is limited and the national average yield is 14.17t·has⁻¹ (SIAP-SAGARPA, 2011), which is considered low, according to the potential yield of 40 t has⁻¹ (Peña and Santiaguillo, 1999). One of the causes of poor performance is the use of native varieties (Santiaguillo et al., 1998). It is reported that the family selection of half-sib and half-sib combined selection are

 Table 1. Mean squares of analysis of variance applied to variables of yield components quality in four diploids and five autotetraploids of husk tomato *Physalis ixocarpa* developed in two locations in Mexico

Maan aguanaa	Sources of variation				
Mean squares	L	Rep. (L)	Р	P x L	Error
FY	0.83**	0.40*	0.21**	0.03ns	0.028
AFW	27.46**	0.86**	5.90ns	0.43ns	0.403
NFP	697.57*	1241.54**	494.59**	431.61**	99.936
EFD	4200.31**	6.09ns	136.28**	58.15**	6.102
PFD	685.80**	8.970*	111.90**	8.85*	2.959
TSS	2.67*	0.48 ns	0.43 ns	0.47 ns	0.324
FF	0.00ns	0.12*	0.77**	0.29**	0.050
pН	0.82**	0.01ns	0.09**	0.04 ns	0.025
VC	0.34ns	1.85*	19.94**	0.10 ns	0.322

L = localities; Rep.(L) = repetitions inside localities; P = populations; FY= fruit yield ; AFW= Average fruit weight; NFP = Number of fruits per plant; EFD = Equatorial fruit diameter; PFD= Polar fruit diameter; TSS = total soluble solids; FF=fruit firmness; VC= Vitamin C; ns = not significant (P > 0.05), * significant (p < 0.05), ** highly significant (p < 0.01).

improvement methods appropriate in genetic improvement of husk tomato (Peña and Marquez, 1990). Stratified visual mass selection has been an efficient method for increasing the yield up to 2.8% per cycle of selection but it is difficult to distinguish the best individuals in the lot selection, so more efficient methods are necessary to be evaluated to determine better options (Peña et al., 2002). This importance is due to the increase in consumption and exports, since this crop is becoming more appreciated in other countries. The yield of tomatillo in Mexico is low, however the study of autopolyploids in tomatillo opens the possibility of starting a new breeding strategy for this species. The autopolyploidy is an inducible biological state characterized by acquisition of more than two sets of chromosomes by means of genome duplication. They originate from either sexual reproduction via 2n gametes or by somatic chromosome doubling, which could increase the variability due to cytoplasmic and citonuclear interactions that contribute to genetic diversity. (Etterson et al., 2007), which can be exploited by plant breeders in species diferent (Aleza et al., 2009; Ollitrault et al., 2008; Omran and Mehammad 2008; Rhodes and Zhang 2000; Nugenet and Adelberg 1995). Autopoliploids are characterized by genomic redundancy and polysomic inheritance, increasing effective population size and should possess increased genome flexibility, allowing them to adapt and persist -in the long run- across heterogeneous landscapes, facilitating the management of artificial selection. Genetic redundancy can enable adaptive divergence of duplicated genes (Parisod et al., 2010). In contrast with diploids in F1 hybrids autopolyploids lacked heterosis, however recombinant hybrid generations often showed a recovery of performance to levels approximating or, sometimes, even exceeding, the parental values (Etterson et al., 2007). Autotetraploids may pass from three possible combinations of two alleles of a diploid to five combinations; besides, it is also possible to develop more vigorous plants (Cubero, 2003) with the potential to increase yields in a species in which hybridization using inbred lines is not possible, due to this Physalis ixocarpa is self-incompatibility (Pandey, 1957). In an earlier work by Robledo-Torres et al. (2011), in which Physalis ixocarpa autotetraploids were successfully formed by colchicine induction from diploid variety Rendidora *Physalis ixocarpa* 2n = 2x = 24 which changed to 2n = 4x =48 autotetraploid, confirmed by cytogenetic and flow cytometry analyses, agronomic evaluation showed phenotypic differences between diploid Rendidora and autotetraploids. Top five in performance and quality of fruit autotetraploids obtained by Robledo-Torres et al. (2011) were selected for comparison with four diploid cultivars with outstanding agronomic traits in the source regions, with the

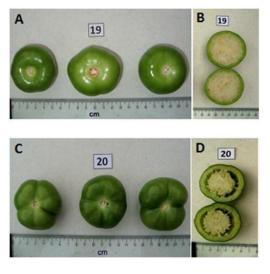


Fig 1. A and B unwrapped fruits, without the calyx, of diploid variety (19D) Rendidora (*Physalis ixocarpa*) showing a total fruit filling. C and D Physalis ixocarpa autotetraploid (20T) fruits showing gaps between the endocarp and mesocarp.

purpose of evaluate them in two locations and compare different autotetraploid with the material from which they originated. The objective of this study was to evaluate and select husk tomato *Physalis ixocarpa* autotetraploid and diploid populations outstanding by yield and quality of fruit for use in future breeding programs for this species.

Results

Fruit yield

The average fruit yield of four diploid populations in General Cepeda (location 1) was of 2.032 kg plant⁻¹ while in Saltillo (location 2) it was of 1.585 kg plant⁻¹. The average values of both localities for diploid populations was 1.809 kg plant⁻¹ with a range of 1.99 kg plant⁻¹ (36.915 t ha⁻¹) to 2.57 kg plant⁻¹ (47.674 t ha⁻¹) corresponding to populations 1D and 13D respectively (Table 2). The fruit yield of autotetraploids in location 1 was 2.00 kg plant⁻¹, and in location 2 of 1.37 kg plant⁻¹. The five tetraploid populations showed an average yield of 1.688 kg plant⁻¹ with a range from 1.43 kg plant⁻¹ (26.54 t ha⁻¹) to 1.92 kg plant⁻¹ (36.59 t ha⁻¹) corresponding to populations 2T and 20T (Table 2). Comparison between diploid and tetraploid, combined variance analysis shows statistically significant differences (p ≤ 0.01) between

Variables Units		Diploid populations					
variables	Units	1D	13D	18D	19D		
FY	kg plant⁻¹	1.99±0.53ab*	2.57±0.65a	1.09±0.39c	1.68±0.63bc		
AFW	g/fruit	46.99±12.91a	43.75±11.24ab	16.06±2.78e	31.17±4.42bcd		
NFP	fruit/plant	44.18±13.15b	60.45±13.28a	66.95±22.15a	53.32±16.50ab		
EFD	mm	46.50±8.07a	43.79±7.53ab	31.80±4.38e	38.93±4.37cd		
PFD	mm	39.91±4.41a	38.42±3.79a	28.65±2.22d	34.14±1.71b		
TSS	°Brix	6.03±0.42a	5.95±0.47 a	6.22±0.67 a	6.13±0.32 a		
FF	kg/cm ²	1.79±0.22ab	1.70±0.21abc	2.01±0.23a	1.65±0.18abc		
pН		4.01±0.16abc	3.93±0.13c	4.14±0.15abc	3.97±0.18bc		
VC	mg/100g	5.53±0.89de	5.92±0.43cde	5.53±0.60de	7.04±0.72b		
		Autotetraploid populations					
		2T	5T	11T	16T	20T	
FY	kg plant ⁻¹	1.43±0.70bc	1.84±0.52ab	1.75±0.47bc	1.49±0.71bc	1.92±0.30ab	
AFW	g/fruit	22.81±8.88ed	30.58±13.60cd	34.99±16.57abc	28.50±14.67cd	29.67±9.00cd	
NFP	fruit/plant	61.74±12.20a	64.31±16.65a	54.41±13.32ab	54.40±16.94ab	69.01±18.36a	
EFD	mm	37.60±9.40b	41.38±10.66bcd	41.43±11.73bcd	40.18±10.26bcd	41.70±10.43bc	
PFD	mm	30.32±5.18cd	31.65±3.35cb	32.03±4.79cb	30.90±3.95cd	32.34±4.21bc	
TSS	°Brix	6.44±0.48 a	6.53±0.64 a	6.18±0.94 a	6.63±0.57 a	6.17±0.84 a	
FF	kg/cm ²	1.09±0.35d	1.70±0.16abc	1.51±0.20bc	1.41±0.27cd	1.08±0.58d	
pН		4.23±0.14a	4.06±0.26abc	4.02±0.16abc	4.12±0.28abc	4.21±0.26ab	
VC	mg/100g	6.50±0.29bcd	9.44±0.90a	10.34±0.51a	5.04±0.53e	6.97±0.69bc	

Table 2. Mean values and standard deviation of yield and fruit quality characteristics in diploids and autotetraploids populations of husk tomato (*Physalis ixocarpa*).

FY= fruit yield; AFW= Average fruit Weight; NFP= Number of fruits per plant; EFD= Equatorial fruit diameter; PFD= Polar fruit diameter; TSS = total soluble solids; FF=fruit firmness; VC= Vitamin C. * Different letters indicate significant differences at (P ≤ 0.05). SD = standard deviation.

locations and between populations, for total fruit yield (Table 1). The average values of FY, obtained in location 1 were higher by 38.74% to those obtained in location 2. That 1D (1.99 kg plant⁻¹) and 13D (2.57 kg plant⁻¹) diploid populations had higher yields of fruit, although they were not statistically different from the 5T (1.8 kg plant⁻¹) and 20T (1.92 kg plant⁻¹) tetraploid populations, which both derived from the diploid population 19D (1.58 kg plant⁻¹) which, in turn, showed a yield of 9.87 to 17.72% lower than populations 5T and 20T. Although the average yield of diploids exceeded in 7.16% the average yield of tetraploids, no statistically significant differences were found (Table 1). However, the average yield of tetraploids was statistically higher than the yield of the population 19D, from which tetraploids were obtained (Table 4). Yields obtained in all populations exceeded the national average value from 87% to 236.39% while yield of the best tetraploid exceeded the national average by 151.18%.

Average fruit weight

In the four diploid populations average fruit weight (location 1) was 28.84 g/fruit while in location 2 was 40.14 g/fruit, and the average weight of fruit in diploid populations was 34.48 g/fruit with ranges from 6.16 g/fruit for 18D diploid to 46.99 g/ fruit for diploid 1D. In autotetraploids, considering both locations, the average fruit weight was 29.31 g/fruit with a range of 22.81 g/fruit for 2T to 34.99 g/fruit 11T population. Comparison of diploid and autotetraploid average fruit weight shows statistically significant differences ($p \le 0.01$) between locations. No differences were presented (P≥0.05), among populations 1D, 13D y 11T, which had the highest AFW (Table 1), although Table 2 shows that the tetraploids had a lowest AFW ($p \le 0.05$) than the diploid Rendidora, because tetraploids had smaller fruits with a partial filling, due to lower pollination. The values of AFW, obtained in location 1 were, 58.8%, above the values obtained in the location 2.

Number of fruits per plant

The number of fruits per plant showed by diploids was 56.22 fruits per plant with values from 44.18 to 66.95 were fruits per plant populations 1D and 18D respectively. In autotetraploids, the NFP was 60.77 fruits per plant with a range of 54.40 to 69.01 fruits per plant for populations 16T and 20T respectively. According to the analysis of variance the variable NFP showed statistical differences between locations ($p \le 0.05$) and between populations ($p \le 0.01$). The NFP value of the tetraploid population 20T resulted statistically higher than population 1D which was the diploid with the highest fruit yield. Table 5 shows that the average number of fruits from tetraploids was statistically higher than the average yield of population 19D which originated the tetraploids. Nevertheless in location 2, NFP was 9.94% higher than in location 1.

Polar and equatorial diameter of fruit

The equatorial diameter of fruit in diploids was 40.25 mm, the smaller diameter, 31.80 mm, was for 18D population, while the wider was 46.50 mm for 1D diploid. The average polar diameter of fruits in diploids was 35.28 mm, with a rank of 28.65 mm for 18D to 39.91 mm for population 1D. The fruit equatorial diameter in autotetraploid was 40.45mm the 2T population having the lowest with 37.60 mm and the best value was for the 20T population with 41.70 mm . For polar diameter autotetraploids showed an average of 31.44 mm and the values ranged from 30.32 to 32.34 mm for 2T and 20T populations. Comparing diploids and autotetraploids for these variables there were significant differences (p \leq 0.01) between locations and between the populations under study (Table 1). For the diploids 1D and 13D DEF they were statistically equal, but the 13D was statistically equal to the autotetraploid 5T, 11T, 16T and 20T. The autotetraploid were statistically superior to the diploids in DEF (Tables 2 and 3).

Table 3. Significance of F values and standard deviation of contrasts designed to compare nine agronomic variables among four diploids and the five autotetraploids of *Physalis ixocarpa*.

Variables	Mean of the four diploids	DS	Mean of the five autotetraploids	DS	F Values
FY	1.808 kg plant ⁻¹	±0.746	1.687 kg plant ¹	±0.567	0.29ns
AFW	34.489 g/fruit	± 15.010	29.311 g/fruit	± 12.868	1.58ns
NF	56.226 fruit/Plant	± 18.028	60.775 fruit/Plant	±15.926	0.05ns
EFD	40.257 mm	± 8.266	40.457 mm	± 10.085	10.76**
PFD	35.281 mm	± 5.400	31.449 mm	± 4.184	8.26**
TSS	6.082°Brix	±0.473	6.390°Brix	±0.702	0.01ns
FF	1.789 kg/cm ²	±0.244	1.359 kg/cm^2	± 0.408	13.16**
pН	4.011	±0.166	4.127	±0.231	1.23ns
VC	6.007mg/100g	±0.896	7.789mg/100g	± 2.066	6.22*

FY= fruit yield per plant; AFW= Average fruit weight; NFP= Number of fruits per plant; EFD= Equatorial diameter; PFD= Polar fruit diameter; TSS = °Brix ; FF=fruit firmness; VC= vitamin C; ns = not significant (P > 0.05), * significant (p < 0.05), ** highly significant (p < 0.01).

Analysis of orthogonal contrast indicates that the tetraploids EFD were statistically higher ($p \le 0.01$) to the diploid 19D Rendidora, therefore doubling the genome, the diameter of fruit in husk tomato increased (Table 4). Diploids 1D and 13D were statistically superior to tetraploids in PFD, the best diploid exceeded by 19% the best tetraploid (Table 3) and diploids had a PFD statistically superior ($p \le 0.01$) totetraploids, likewise, when comparing PFD of population 19D with the PFD of tetraploids, these showed a statistically significant reduction ($p \le 0.01$). Further analysis of variables EFD and PFD shows that when calculating the ratio DEF/DPF diploids showed values from 1.11 to 1.16 while the rank found in tetraploids was from 1.24 to 1.30.

Total soluble solids

For diploid total soluble solids the average value was 8.6 °Brix and the values were 5.95 °Brix for population 13D and 22.6 °Brix for 18D. Autotetraploid populations showed mean values of total soluble solids of 6.39 °Brix within a range of 6.17 to 6.63 °Brix for 20T and 16T respectively. Statistically significant differences ($p \le 0.01$) were found between locations 1 and 2 for total soluble solids, however for SST there were no significant differences between populations (Table 1).

Firmness of fruit

In fruit firmness diploids presented 1.78 kg/cm² with a range of 1.65 to 2.01 kg/cm² for populations 19D and 18D. For autotetraploid populations fruit firmness was 1.35 kg/cm² and values ranged from 1.08 to 1.41 kg/cm² for 20T and 16T populations. The analysis of variance showed statistically significant differences between locations for fruit firmness, therefore this variable was the most stable one across environments. However, a wide variability was found in FF, showing statistically significant differences ($p \le 0.05$) between populations (Table 2). Among the populations under study a wide variability in FF was found which is shown by statistically significant differences ($p \le 0.05$) between them statistically, diploids were higher in FF than tetraploids (Table 2). Firmness of the fruit was negatively modified by genome duplication since the FF value in population 19D was 18% higher than values observed in their derived tetraploid populations (Table 4).

pН

For this variable diploids presented an average of 4.01, and values ranged from 3.93 to 4.14 for the 13D and 18D populations respectively. In autotetraploids there was an average value of 4.12 with a minimum value of 4.02 (11T)

and a maximum of 4.23 for 2T population. Statistically significant differences ($p \le 0.01$) in the pH of the fruit, indicate the great variability among the populations studied, therefore wide variability in taste, although this feature is not influenced by changes in the ploidy level. It is also possible to obtain fruits with differences in taste, although this feature is not influenced by changes in the ploidy level.

Vitamin C contents

It was found that the average content of vitamin C of the diploids was of 6.00 mg/100g and ranged from 5.53 to 7.04mg/100g corresponding to populations 18D and 19D, respectively. Vitamin C in autotetraploids presented an average value of 7.65 mg/100g ranging from 5.04 mg/100g in population 16T to 10.34 mg/100g in autotetraploid 11T. There were statistically significant differences ($p \le 0.01$) between locations and between populations for vitamin C contents, indicating that populations had a different behavior in localities, and that there were differences between the populations under study (Table 1). The fruits of autotetraploid population 11T (10.33mg/100g) showed the highest content of VC, significantly exceeding the diploid 13D (5.92mg/100g) by 69.7%.

Discussion

Fruit yield

Statistical differences observed between sites allow deducing that the evaluated populations have specific adaptation traits to studied environments. In this sense, Blum (1988) indicates that the factors responsible for performance across contrasting environments are relevant to understanding the nature of yield stability and may open the way for the development of additional selection criteria for yield of fruit. Given the statistical differences observed between the populations under study it may be concluded that there is a wide diversity among both diploid and tetraploid populations as a result of cross-pollination owing partially to the selfincompatibility existing in the species (Pandey, 1957). In this study significant differences were found in fruit yield per plant, between diploid Rendidora and autotetraploids, as opposed to the reported by Robledo-Torres et al. (2011).On the other hand, polyploidy significantly increases the genetic variability since two alleles of a gene in a diploid are able to make three combinations, but in a tetraploid it is possible to form five combinations with the same number of alleles (Cubero, 2003). Thus, considering the whole set of genes existing in the 24 chromosomes of the husk tomato, variability may be significantly increased. Besides, the geographically distant origin between the specimens used in

Table 4. Significance of F values and standard deviation of contrasts designed to compare nine agronomic variables between diploid variety *Rendidora* and the five autotetraploids of husk tomato.

Variables	Mean of Diploid Rendidora (19D)	DS	Mean of the five autotetraploids	DS	F Values
FY	1.581 kg plant ⁻¹	±0.49	1.687 kg plant ¹	±0.567	16.21*
AFW	31.165 g/fruit	± 4.42	29.311 g/fruit	± 12.868	41.48**
NFP	53.321 fruit/Plant	± 16.50	60.775 fruit/Plant	±15.926	4.75*
EFD	38.934 mm	±4.37	40.457 mm	± 10.085	96.89**
PFD	34.139 mm	± 1.71	31.449 mm	± 4.184	56.76**
TSS	6.133°Brix	±0.32	6.390°Brix	±0.702	0.03ns
FF	1.654kg/cm ²	±0.18	1.359 kg/cm^2	± 0.408	38.91**
pН	3.966	±0.18	4.127	±0.231	0.49ns
VC	7.041 mg/100g	±0.72	7.789 mg/100g	± 2.066	37.88**

FY= fruit yield per plant; AFW= Average fruit weight; NFP = Number of fruits per plant; TSS = $^{\circ}$ Brix ; EFD = Equatorial fruit diameter; PFD= Polar fruit diameter; VC= Vitamin C; ns = not significant (P>0.05), * significant (p<0.05), ** highly significant (p<0.01)

Table 5. Diploid and autotetraploid used populations.				
Key	Diploid	Origin		
	Populations			
1D	Landraces Felipe Ángeles	Acatzingo, Puebla		
13D	Gran Esmeralda	Empresa Harris Moran		
18D	Landraces Morado Tamazula	Arandas, Jalisco		
19D	Rendidora	Zacatepec, Morelos		
Key	Autotetraploid	Origin		
	Populations			
2T 5T 11T 16T 20T	UAN-II-113 UAN-III-16 UAN-III-7 UAN-II-107 UAN-II-16	Artificially formed from the variety Rendidora by the action of colchicine (Robledo-Torres., et al 2011).		

this research is considered as another source of variability since biotic and abiotic factors in each region are specific to each one of them. Since the average yield of tetraploids was statistically higher than diploid population 19D, from which tetraploids were obtained (Table 4), it can be deduced that autotetraploid establishment may be a promising method for increasing the fruit yield in husk tomato crops. According to Molero and Matos (2008), chromosomal duplication in polyploid cells causes an increase in plant height and length, width, thickness, and leaf volume with respect to diploid and chimera plants, It is considered that polyploidization could be an alternative for increasing cell biomass production in Physalis ixocarpa. The best tetraploid yields exceeded the national average yield of 14.17 t ha -1 (SIAP-SAGARPA, 2011). For this reason it is considered that these new tetraploids have a high potential to develop new hybrid lines, or to be used as parents in triploid hybrid production, as has happened in other crops such as melon, where high yields have been achieved using tetraploids (Nugent and Ray, 1992; Batra, 1952; Dumas, 1974). It is noteworthy that the evaluated autoteraploids should undergo a process of selection and stabilization in order to achieve the full expression of their genetic potential. According to He et al. (2011) selection should be done on the basis of the meiotic stability of tetraploids, since it has been found that using parent lines with meiotic stability improves embryo development, and seed production rate of tetraploid hybrid rice.

Average fruit weight

The lower AFW in tetraploids is due, in part, to the fact that even though tetraploids showed a higher number of fruits they were smaller and, in some cases, empty gaps between the fruit mesocarp and endocarp was observed. This fact may be the consequence of poor pollination resulting in a low seed formation and, therefore, a low synthesis of gibberellins and auxins, growth regulators promote cell division and development of fruits (Bünger and Bangerth, 1982). The low pollination efficiency is the result of meiotic irregularities that lead to loss of chromosomes at anaphases I and II, producing extra micronuclei and abnormal pollen grains (Cequea, 2000) leading the plant to a reduced seed production or loss of fertility. It is a known fact that the new autopolyploids are highly variable in this feature (Ramsey and Schemske, 2002) unlike diploids that normally have a regular meiosis resulting in eggs and pollen grains without chromosomal irregularities which produce normal fruits (Fig 1). Meiotic analysis in diakinesis and metaphase I showed bivalent pairing for the Rendidora cultivar, while the autotetraploid plants exhibited pairing irregularities, with the presence univalents, bivalents, trivalents and quadrivalents (Robledo-Torres et al., 2011).

Number of fruits per plant

The higher number of fruits in tetraploids population 20T may be due to the higher vegetative growth of the plants. As higher amount of biomass is produced the demand for nutrients is also higher, the plants are taller and need more days for flowering and harvest (Robledo-Torres et al., 2011); they also have a broader ecological tolerance, and larger cells (Cequea, 2000).

Polar and equatorial diameter of fruit

A comparison of population 19D PFD with the PFD of tetraploids leads to the conclusion that the increase in ploidy

level induced a reduction of this variable. Thus, duplication of genome induced a reduction in the PFD. In autotetraploid was increasing the fruit equatorial diameter and reduced the polar diameter of fruit which indicate that doubling the ploidy level changed the fruit's shape since the obtained fruits were flattened at the poles (Fig 1). Similar results were reported in tetraploid melons (Nugent and Adelberg, 1995).

Total soluble solids

As far as TSS content in husk tomato fruit is concerned, there were no differences among diploids and tetraploids, therefore increasing the ploidy level did not change the plant's ability to synthesize soluble solids (sugars, organic acids, vitamins, and amino acids), but it was found that all assayed materials yielded higher values than the ones reported in husk tomato by Santiaguillo et al. (2004). These results agree with Nugent and Adelberg (1995) who found that hybrid melon triploids showed higher sugar levels than their diploid progenitors.

Fruit firmness

The low fruit firmness in tetraploids was due to some partial filling which results in a gap have been demonstrated between the endocarp and mesocarp. This negative trait should be avoided during the selection process for tetraploids generation. In order to improve this negative trait in autotetraploids, it is necessary to assay alternatives as the ones used by Qiu et al. (1995) who delayed tissue softening by increasing calcium levels in papaya fruit keeping, this way, the quality thereof. These results could be related to the findings of Tomekpe et al. (2004) in banana fruit, where the loss of firmness was due to the high water content in tetraploid fruits that quickly ripened and softened. Furthermore, according to Ospina et al. (2007) the condition of fruit at harvest time, temperature, and storage conditions may have influenced the yields.

pН

Although with increasing ploidy level in husk tomato no significant differences were found in the pH value in the fruits, this value remained within the normal range, therefore the pH values found in these tomatoes make both, diploid and tetraploid, to stay within the range of good quality, since they were acid.

Vitamin C contents

In the vitamin C content autoteraploids were higher than diploids, there were even some of them who doubled the value of the diploids, perhaps because polyploids are more capable of producing secondary metabolites (Cequea, 2000), so these results indicate that genome duplication in husk tomato could increase the vitamin C content, a feature that could be relevant, because vitamin C is one of the main nutrients in fruits and vegetables whose contribution to human diet depends exclusively on these sources, besides vitamin C has an important role in human nutrition because it is an antioxidant that contributes to human health and is credited with strengthening the body in defense of cardiovascular diseases (Carr and Frei, 1999). These preliminary results indicate that Physalis ixocarpa autotetraploids can be used to obtain improved genotypes with antioxidant properties. In a similar way Viehmannová et al. (2012) found that by induction of polyploidy in Ullucus tuberosus microtubers the octaploids average weight was lower as compared with diploids, however, these were high in vitamin C and also had a lower starch content. This is a new contribution since the content of vitamin C was not evaluated by Robledo-Torres et al. (2011).

Materials and methods

Plant materials

Four diploid and five autotetraploids populations were used. The diploids used were: (19D) Variety Rendidora, (13D) Grand Esmeralda, landraces (1D) Felipe Angeles, and (18D) Purple Tamazula. Landraces are materials with outstanding performance in the areas of collection Table 5 description. The autotetraploids were developed by Robledo-Torres et al. (2011) using the two colchicine concentrations 0.12% and 0.16%, 38 polyploid plants were obtained the Physalis ixocarpa. From their bulked progeny of 1800 plants, 15 outstanding polyploids were selected. From among the autotetraploid populations evaluated by Robledo-Torres et al. (2011) the best five autotetraploid populations were selected 2T, 5T, 11T, 16T, 20T (Table 5), on the basis of yield an fruit quality. Agronomic evaluation of the above mentioned material was held in 2010 in two locations in Mexico: Location 1 General Cepeda, Coah., Mexico (1480 m, 25 ° 22 '21" LN, 101 ° 28' 8" LO). Annual average temperature: 17.6 ° C, annual average highest temperature: 25.3 ° C, annual average lowest temperature: 10.3 ° C, average annual humidity: 53.8%. Total cumulative annual rainfall: 285.4 mm. Location 2 Saltillo, Coah., Mexico (1742 m, 25 ° 14 '15" LN, 101 ° 10' 16" LO), average annual temperature: 15.5 ° C. Annual average highest temperature: 23.2 ° C. Annual average lowest temperature: 8.4 ° C, average annual humidity: 53.7%, total cumulative annual rainfall: 718.6 mm (SAGARPA-INIFAP, 2012)

Crop management

The populations were planted in polystyrene trays with 200 cavities placing two to three seeds per hole using peat and perlite as substrate at a ratio of 1:1. Seedlings were grown under greenhouse conditions and taken to the field for transplanting after reaching 10 to 12 cm in height and two pairs of true leaves appeared. Field transplantation was performed in May 2010 in the two experimental locations. The plants were established in growing beds 1.8 m wide, two rows per bed, at a distance of 45 cm between rows and 60 cm between plants within the row. Thinning was performed ten days after transplantation leaving only one seedling per point. The nine populations were established under an experimental randomized block design with four replications, using ten plants as experimental unit. Daily drip irrigation was applied at a rate of 1L h⁻¹ for three hours. Nutrition was administered via irrigation three times a week in the first 9 weeks, applying N, P, K, Ca, and Mg solution in amounts of 2.40, 1.44, 1.47, 0.39 and 0.37 Kg ha⁻¹ respectively. Later on, it was changed to 1.90, 1.85, 2.75, 0.37 and 0.47 Kg ha⁻¹(N, P, K, Ca, and Mg) until week 15. Since there were pest problems with Diabrótica (Diabrotica spp.) throughout the crop cycle applications of Endosulfan and Methomyl were necessary every other week.

Yield measurement and its components

The evaluated variables in the two localities were: Fruit yield (FY) in Kg plant⁻¹ determined by weighing all of the fruits produced per plant at harvest time, average fruit weight

(AFW) in grams, and total number of fruits per plant (NFP). The three variables were estimated from a random sample of 10 plants in each of four replicates in five cuts at intervals of 8 days. The polar fruit diameter (PFD) and equatorial fruit diameter (EFD) values were measured with a digital precision caliper (AutoTECTM), in three fruits per plant from three plants chosen at random from each population with repetitions at the second, third and fourth cut.

Fruit quality measurement

The total soluble solids (TSS) were estimated in fruit juice with an Atago N-1E® refractometer and expressed in degrees (°Brix). The firmness was determined with a penetrometer trademark Fruit Presure Tester, model FT-327 of 13 kg, with 8 mm tip, removing the cuticle of each fruit at two opposite points of the equatorial and the penetrometer tip was introduced up to the mark with a single push taking two readings per fruit and reported in kg/cm². The pH values were obtained from fresh fruit juice concentrates and with a digital Corning® potentiometer model 320. Vitamin C was determined according to the official AOAC (2000) methodology in mg/100g. All these variables were measured in three fruits per plant from three plants taken at random from each population with repetitions at the second, third and fourth cut, which were made at 84, 93, and 100 days respectively after transplantation.

Statistical analysis

Combined analysis of variance and Tukey's mean values comparisons ($P \le 0.05$) were applied. Likewise, orthogonal contrast analysis using SAS statistical software ver. 9.0 was also executed (SAS, 2008).

Conclusions

The diversity in husk tomato diploid populations can be significantly increased with the development of autotetraploids which may then be used to carry out a proper selection process and develop populations with higher yields by taking advance of their higher genetic variability. On the other hand, autopolyploid formation in husk tomato can increase the content of vitamin C and improve other characteristics related to fruit quality. However, chromosomal duplication in husk tomato unfortunately contributed to undesired changes in the shape of fruits which became flattened at the poles with gaps between the endocarp and mesocarp, features that could be improved by selecting plant specimens with meiotic pairing stability that allow the development of uniform and productive cultivars.

Acknowledgements

To the Universidad Autónoma Agraria Antonio Narro (UAAAN). the Universidad Autónoma de Nuevo León (UANL, FCB) and to the National Council for Science and Technology (CONACYT).

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