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Genetic divergence among eggplant genotypes under high temperatures

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ABSTRACT

The aim of this study was to estimate the genetic divergence among eggplant genotypes for agronomic traits in order to gather information for the selection of genotypes in eggplant breeding programs for tolerance to high temperatures. Ten traits recommended by the International Board for Plant Genetic Resources were analyzed in 24 genotypes, arranged in a randomized complete block design with four replicates. Data were submitted to analysis of variance ($P < 0.01$) and later to the UPGMA and Tocher grouping methods, using the generalized Mahalanobis distance (D^2) as dissimilarity measure. Three and six groups of similarity were obtained, respectively, for the multivariate techniques used, UPGMA and Tocher, with concordance in the grouping of 87.50% of the genotypes. The characters fruit length (34.71%), fruit width (35.96%) and fruit length/width ratio (14.08%) were the main contributors to genetic divergence, explaining 90.72% of total genetic dissimilarity. The genotypes presented considerable genetic variability for all agronomic traits analyzed and can be used in eggplant genetic breeding programs for high temperatures.

Keywords: *Solanum melongena*, genetic variability, protected cultivation.

RESUMO

Divergência genética entre genótipos de berinjela sob altas temperaturas

O objetivo deste trabalho foi estimar a divergência genética entre genótipos de berinjela para caracteres agrônômicos, visando gerar informações para a escolha de genótipos em programas de melhoramento genético para tolerância a altas temperaturas. Foram analisados dez caracteres recomendados pelo International Board for Plant Genetic Resources em 24 genótipos, dispostos no delineamento de blocos ao acaso, com quatro repetições. Os dados foram submetidos à análise de variância ($P < 0,01$) e posteriormente aos métodos de agrupamento de UPGMA e Tocher, utilizando-se a distância generalizada de Mahalanobis (D^2) como medida de dissimilaridade. Obtiveram-se três e seis grupos de similaridade, respectivamente, para as técnicas multivariadas utilizadas, UPGMA e Tocher, havendo concordância no agrupamento de 87,50% dos genótipos. Comprimento do fruto (34,71%), largura do fruto (35,96%) e a relação comprimento/largura do fruto (14,08%) foram os caracteres que mais contribuíram para a divergência genética, explicando 90,72% da dissimilaridade genética total. Os genótipos apresentaram considerável variabilidade genética para todos os caracteres agrônômicos analisados e podem ser utilizados nos programas de melhoramento genético de berinjela para altas temperaturas.

Palavras-chave: *Solanum melongena*, variabilidade genética, cultivo protegido, correlações genéticas.

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In Brazil, the area cultivated with eggplant (1550 ha/year) is concentrated mainly in the Center-South region (Boiteux *et al.*, 2016). In the Northeast, where temperatures are relatively high, averaging around 28°C and peaking around 40°C (Ramalho, 2013) crop yields have been unpredictable. This is mainly due to flowering coinciding with warmer periods of the year, increasing the occurrence of malformation and/or fruit abortion. In greenhouse crops, where the internal temperatures are higher than the outside, there is a considerable reduction in crop yield in the region (Valadares *et*

al., 2019ab).

The optimal temperature for crop growth and development is in the range of 22 to 30°C (Adamczewska-Sowińska & Krygier, 2013). When the temperature exceeds 32°C, productivity is drastically reduced (Baswana *et al.*, 2006). Adoption of strategies for evaluation and selection of eggplant genotypes and knowledge of the genetic variability involved in traits of agronomic importance are extremely important for the choice of genotypes to compose eggplant breeding programs for high temperature tolerance.

Genetic divergence studies provide these parameters and allow the correct

choice of parents which, when crossed, result in high heterotic effect on progenies, maximizing the chances of obtaining superior genotypes in segregating generations (Rotili *et al.*, 2012). These genotypes can be obtained by biometric techniques based on quantification of heterosis or by predictive processes (Nardino *et al.*, 2017).

Among the biometric techniques are diallel analyzes, which generate information about the specific combining ability and heterosis manifested in hybrids and in the prediction of genetic divergence, also keeping in mind

that several multivariate methods can be applied, including agglomerative methods.

Agglomerative methods (Cruz *et al.*, 2012) seek to genetically discriminate individuals and allow them to be separated into groups by analyzing a set of characters inherent to each individual, grouping them by some classification criteria, so that there is homogeneity within each group and heterogeneity between them. They also basically involve two stages, the first refers to the estimation of a similarity or dissimilarity measure and the second refers to the adoption of a grouping technique.

As dissimilarity measures, we can point out the Euclidean distance, the average Euclidean distance, the average squared Euclidean distance, the weighted distance and the generalized Mahalanobis distance (D^2) (Cruz *et al.*, 2012, 2014).

Genotype grouping can be done by optimization and hierarchical clustering methods. Among the optimization clustering methods are the modified Tocher and Tocher (Vasconcelos *et al.*, 2007; Cruz *et al.*, 2014). Hierarchical clustering methods include the methods of the nearest neighbor, the farthest neighbor, UPGMA (Unweighted Pair-Group Method using Arithmetic Averages), the centroid, the median (or WPGMC), and the Ward's minimum variance (Cruz *et al.*, 2012).

Finally, we can adopt the cophenetic correlation analysis to increase the reliability of the conclusions regarding interpretation based on dendrograms. This establishes a correlation between the similarity or dissimilarity matrix with the generated dendrogram, i.e., compares the actual distances obtained between the accessions with the distances graphically represented (Kopp *et al.*, 2007). The higher the correlation value, the smaller the distortion caused by grouping.

Given the above, the present work aimed to estimate genetic divergence between eggplant genotypes for agronomic traits, aiming to generate information for the choice of genotypes in eggplant breeding programs for high temperature tolerance.

MATERIAL AND METHODS

The experiment was conducted between May and September 2016 at Universidade Federal Rural de Pernambuco (UFRPE), Recife-PE.

Seeds were sown in 128-cell expanded polystyrene trays filled with inert substrate (sifted coconut powder). Trays were kept in greenhouse in the hydroponic system by sub-irrigation until reaching the point for transplantation, plantlets with three definite leaves. Seedlings were individually transplanted to 5 L pots, containing inert substrate (coconut powder), spaced 1.75 m between rows and 0.60 m between plants.

Plants were cultivated in open hydroponics with substrate, under a 30 m long, 14 m wide, 3 m ceiling height arch, with 50% shading side screens and roof covered with a low-density polyethylene film, 150 micrometers thick.

Mineral nutrition and water requirement of plants were supplied by balanced nutrient solution at each plant development stage. A drip irrigation system was used with 2 L h⁻¹ emitter, automatically controlled by a digital timer, with irrigation amounts and duration adjusted according to environmental conditions of the region and the amount of nutrient solution absorbed by the plants.

Throughout the experiment period, relative air temperature (average, maximum and minimum) and relative air humidity were recorded using a HOBO mini datalogger. The environmental conditions in which the experiment was performed are characterized by high temperatures, since in all phenological phases temperatures exceeded the optimum range of the culture.

Eighteen eggplant accessions from the Embrapa Hortaliças' germplasm bank and six commercial cultivars (Ciça F1, Choryoku F1, Kokushi Onaga F1, Ajimurasaki F1, Ajishirakawa F1 and Florida Market) were evaluated, coming to a total of 24 treatments arranged in randomized block design with four replications and four plants per experimental plot.

Six quantitative traits were evaluated: fruit length (cm), fruit width (cm), fruit length/width ratio, number of fruits per plant, yield per plant (g) and fruit mass (g); and four qualitative traits: fruit color at commercial maturity (1= green; 2= white; 3= yellow; 4= light red; 5= dark red; 6= grayish purple; 7= purple; 8= dark purple; 9= black), fruit color distribution at commercial maturity (1= uniform; 3= mottled; 5= lacy; 7= streaked), fruit curvature (1= none (straight fruit); 3= slightly curved; 5= curved; 7= snake-shaped; 8= sickle-shaped; 9= U-shaped) and the presence of thorns in the fruit's cup (0= none; 1= very few (<3); 3= few (~5); 5= intermediate (~10); 7= many (~20); 9= very many (>30)) (IBPGR, 1990).

Quantitative data were initially submitted to univariate analysis of variance ($p < 0.01$) and from the means and residual variance and covariance matrix was obtained the genetic dissimilarity matrix based on the generalized Mahalanobis distance (D^2). The genotype clustering was obtained by the method of ascending hierarchical classification algorithm UPGMA (Unweighted Pair-Grouped Method Average) and by the Tocher's optimization method.

The relative importance of traits in the prediction of genetic diversity was also studied through the participation of D^2 components, related to each trait in the total dissimilarity observed, and the diversity between genotypes was estimated by Mahalanobis distance. (Singh, 1981).

To test the efficiency of the hierarchical clustering method, we estimated the cophenetic correlation coefficient, obtained with 1,000 simulations, analyzed by the "t" test. The cutoff point (C_p) of the dendrogram formed by the UPGMA method was defined as proposed by Mojema (1977), following the formula $C_p = m + ksd$, where m = the mean distance values of the fusion levels corresponding to the stadiums; $k = 1.25$ (Milligan & Cooper, 1985); sd = standard deviation.

All statistical analyzes were performed using the GENES software, version 1990.2018.75 (Cruz, 2013).

RESULTS AND DISCUSSION

The micrometeorological data obtained during the experiment period showed that the maximum air temperature in the greenhouse ranged between 29.8 and 41.4°C and the minimum temperature between 18.6 and 23.7°C. The average temperature ranged between 23.7 and 28.5°C. Thus, the environment was classified as high temperature for eggplant cultivation. Relative humidity ranged from 83.7 to 95.4%.

Significant differences were verified by F test ($p < 0.01$) between genotypes for all analyzed traits (Table 1). This result refers to the existence of phenotypic variability between genotypes, and it is necessary to identify the superior genotypes to be crossed in eggplant

breeding programs.

Dissimilarities (D^2) between genotypes ranged from 1.07 to 728.53, with an average of 133.47. The largest distances were recorded between CNPH 135 and Ajishirakawa F1 genotypes. On the other hand, genotypes CNPH 47 and Florida Market were the least genetically distant (Figure 1). Thus, crossings between the most divergent groups are indicated for formation of segregating populations and with greater genetic variability for the analyzed traits.

The dendrogram obtained by UPGMA hierarchical method showed the formation of three groups, considering a significant cut of 44.32% (Mojena, 1977). Group 1 was composed of most genotypes, approximately 84% (Figure 1). Among quantitative traits, those

that most contributed to the genetic divergence stood out (Table 3). In this sense, the fruits of this group had an average length of 14.11 cm, with averages ranging from 6.89 (CNPH 668) to 18.09 cm (CNPH 51). For fruit width, the average was 5.84 cm, with values between 3.64 (CNPH 84) and 8.55 cm (CNPH 135), reflecting in the length/width ratio of the fruit, which was between 1.48 (CNPH 668) and 4.91 (CNPH 84) with a mean of 2.59 (Table 1). Results similar to those were reported by Valadares *et al.* (2019b).

In the morphological description of the genotypes of group 1, for qualitative traits (Table 1), considerable levels of phenotypic variability were observed only for fruit color at commercial maturity, with a predominance of dark purple, followed by grayish purple,

Table 1. Description of eggplant genotypes under high temperatures. Recife, UFRPE, 2016.

Genotypes	LF (cm)	FW (cm)	FLWR	NFP	YP (g)
CNPH 135	12.96	8.56	1.52	1.89	470.03
CNPH 60	13.51	6.77	2.00	2.14	301.03
CNHP 51	18.09	5.00	3.67	1.57	203.20
CNPH 410	15.29	5.48	2.80	1.39	149.72
CNPH 84	17.70	3.64	4.91	3.56	283.50
CNPH 71	16.01	4.09	3.94	1.85	161.08
CNPH 668	6.89	4.67	1.48	4.08	209.79
CNPH 146	12.63	5.85	2.17	2.35	249.95
CNPH 140	13.42	5.27	2.83	2.09	191.20
CNPH 93	13.82	4.61	2.99	1.97	173.23
CNPH 47	14.15	7.68	1.85	1.26	242.36
CNPH 141	11.56	5.86	1.97	3.83	383.69
CNPH 67	13.06	6.38	2.05	3.00	341.20
CNPH 107	15.56	5.58	2.80	2.24	317.69
CNPH 53	11.90	7.77	1.55	1.00	151.28
CNPH 109	14.47	6.18	2.34	1.72	225.65
CNPH 79	13.16	5.33	2.60	2.00	101.41
Ciça F1	16.59	6.21	2.68	1.42	242.56
CNPH 100	17.35	4.56	3.91	1.16	110.99
Florida Market	14.26	7.50	1.91	1.39	257.40
Ajishirakawa F1	23.65	3.21	7.41	1.30	130.94
Choryoku F1	30.02	3.99	7.51	1.13	154.54
Kokushi Onaga F1	27.95	4.42	6.39	1.67	223.60
Ajimurasaki F1	28.36	2.84	9.97	4.13	316.13
CV (%)	7.58	6.50	10.86	28.78	32.29
QM _(treatments)	128.83**	8.83**	19.82**	3.65**	32586.12**
Mean	16.35	5.48	3.47	2.08	233.01

Table 1. Continuation

Genotypes	FM (g)	FCCM	DFCCM	FC	TFC
CNPH 135	248.16	dark purple	uniform	none (straight fruit)	intermediate
CNPH 60	139.12	dark purple	uniform	none (straight fruit)	none
CNHP 51	127.49	dark purple	uniform	slightly curved	none
CNPH 410	105.68	purple	uniform	none (straight fruit)	none
CNPH 84	77.69	purple	uniform	curved	none
CNPH 71	89.06	purple	streaked	curved	none
CNPH 668	51.47	green	streaked	none (straight fruit)	none
CNPH 146	108.66	grayish purple	uniform	none (straight fruit)	intermediate
CNPH 140	91.53	grayish purple	uniform	none (straight fruit)	few
CNPH 93	77.35	grayish purple	uniform	none (straight fruit)	none
CNPH 47	174.82	dark purple	uniform	none (straight fruit)	none
CNPH 141	98.41	dark purple	uniform	none (straight fruit)	few
CNPH 67	111.23	grayish purple	uniform	none (straight fruit)	none
CNPH 107	140.49	dark purple	uniform	none (straight fruit)	none
CNPH 53	167.53	dark purple	uniform	none (straight fruit)	none
CNPH 109	134.09	purple	uniform	none (straight fruit)	none
CNPH 79	47.84	grayish purple	uniform	none (straight fruit)	none
Ciça F1	168.41	dark purple	uniform	none (straight fruit)	none
CNPH 100	113.39	dark purple	uniform	none (straight fruit)	none
Florida Market	187.18	dark purple	uniform	none (straight fruit)	none
Ajishirakawa F1	101.27	white	uniform	curved	none
Choryoku F1	138.04	green	uniform	curved	none
Kokushi Onaga F1	142.96	black	uniform	snake-shaped	none
Ajimurasaki F1	77.68	purple	uniform	snake-shaped	none
CV (%)	24.60	-	-	-	-
QM _(treatments)	8472.63**	-	-	-	-
Mean	121.65	-	-	-	-

LF= fruit length; FW= fruit width; FLWR= fruit length/width ratio; NFP= fruits per plant (number); YP= yield per plant; FM= fruit mass; FCCM= fruit color in commercial maturation; DFCCM= distribution of fruit color during commercial maturation; FC= fruit curvature; TFC= thorn in the fruit cup; **significant at 1% probability by F test.

purple and green. However, the fruits showed color distribution at commercial maturation predominantly uniform with no curvature and no thorns in the fruit's cup (Table 1). This distribution indicates that, in relation to the evaluated traits (quantitative and qualitative), most genotypes presented high levels of similarity, including the commercial cultivars Ciça F1 and Florida Market, contemplated in this group 1.

According to Guedes *et al.* (2013), individuals are grouped in pairs, using arithmetic means of dissimilarity, and the dendrogram prioritizes genotypes with greater similarity. This explains why the Kokushi Onaga F1, Ajishirakawa F1 and Choryoku F1 genotypes formed group 2 and the Ajimurasaki F1 genotype

alone group 3, consisting of fruits longer than 23.64 cm, fruit width less than 4.41 cm and length/width ratio of the fruit greater than 6.38 (Table 1). Averages for fruit length in group 2 were between 23.65 (Ajishirakawa F1) and 30.01 cm (Choryoku F1) and for fruit width between 3.20 (Ajishirakawa F1) to 4.41 cm (Kokushi Onaga F1). For length/width ratio of the fruit, the variation ranged from 1.48 (CNPH 668) to 4.91 (CNPH 84) (Table 1). For fruit color, Ajishirakawa F1 genotype presented white, Choryoku F1 green and Kokushi Onaga F1, black fruits. However, predominantly of uniform distribution and without any thorn in the fruit's cup. About fruit curvature, Ajishirakawa F1 and Choryoku F1

genotypes presented curved fruits and Kokushi Onaga F1 snake-shaped fruits (Table 1). No non-commercial genotype showed considerable similarity with these commercial cultivars.

Group 3 included only the Ajimurasaki F1 genotype with the second longest fruit length among the evaluated genotypes (28.35 cm), smallest fruit width (2.83 cm) and highest fruit length/width ratio (9.96) (Table 1), similar to those reported by Valadares *et al.* (2019b). The fruits showed uniform purple coloration, snake-shaped curvature and no thorns in the fruit cup (Table 1).

Grouping of genotypes by the Tocher method was partially similar to the UPGMA method when grouping

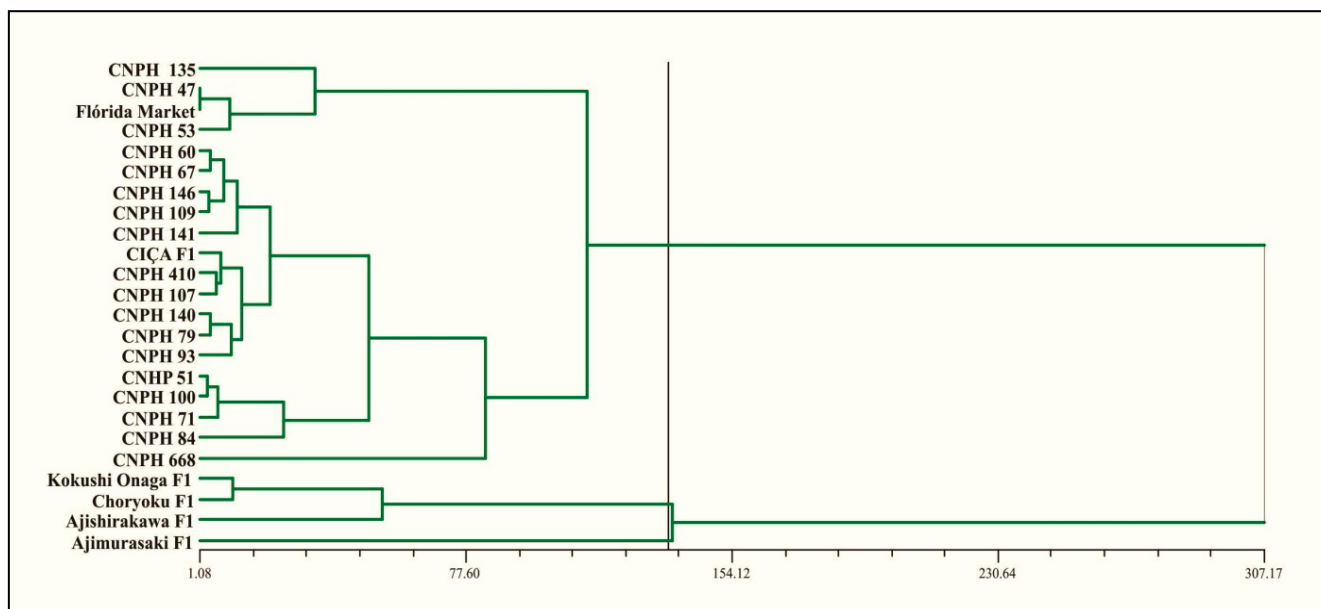


Figure 1. Dendrogram obtained by UPGMA grouping method, using Mahalanobis distance (D^2), resulting from the analysis of 24 eggplant genotypes, evaluated under high temperatures. Recife, UFRPE, 2016.

Table 2. Grouping by Tocher method resulting from the analysis of 24 eggplant genotypes evaluated under high temperatures. Recife, UFRPE, 2016.

Groups	Genotypes
1	CNPH 47, Flórida Market, CNPH 53, CNPH 60, CNPH 67, CNPH 109, CNPH 146, CNPH 141, Cİça F1, CNPH 140, CNPH 107, CNPH 79, CNPH 410, CNPH 93, CNPH 51, CNPH 100, CNPH 71
2	AjishirakawaF1, ChoryokuF1, Kokushi Onaga F1
3	CNPH 84
4	Ajimurasaki F1
5	CNPH 668
6	CNPH 135

among the most divergent genotypes (Table 2). Similarity between the different clustering techniques can be seen from the fact that genotypes belonging to Tocher's group 1 were mostly the same ones from the UPGMA grouping, around 71% of the genotypes, including Cİça F1 and Florida Market.

There was also agreement in the formation of group 2 which included genotypes Kokushi Onaga F1, Ajishirakawa F1 and Choryoku F1 and the formation of group 4 composed only by genotype Ajimurasaki F1. Agreement between multivariate techniques is important in the study of genetic divergence, as it allows the recommendation of crossing between the most divergent parents possible, in order to broaden the genetic base and

consequently increase genetic variability (Abreu *et al.*, 2004). Disagreements occurred in the formation of groups 3 (CNPH 84), 5 (CNPH 668) and 6 (CNPH 135) by Tocher's method.

The association of clustering techniques provides a more efficient support for determination of divergence, since Tocher discriminates each group and UPGMA discriminates each genotype and can more safely infer the use of parents in breeding programs (Bertan *et al.*, 2006).

The relative importance of the analyzed traits in the genetic dissimilarity between genotypes was detected by Singh's method (1981). This method considers that the most important characteristics express greater variability. In this respect, we found

that fruit length, fruit width and fruit length/width ratio presented the highest percentage of contribution to divergence among the 24 evaluated genotypes, explaining 90.72% of the total genetic dissimilarity (Table 3).

High contribution of fruit length to eggplant divergence has been reported by Babu & Patil (2004) and Mehta *et al.* (2004), while average fruit weight and number of fruits per plant traits have lower contributions as reported by Prabakaran *et al.* (2015). Bashar *et al.* (2016) also cited contributions of length and width of fruit traits in the genetic divergence of eggplant. we observed that genotype clustering was predominantly influenced by fruit length, fruit width and fruit length/width ratio, showing greater variability for

Table 3. Relative contribution of six quantitative traits to genetic divergence among 24 eggplant genotypes, using the Singh method, evaluated in 24 eggplant genotypes under high temperatures. Recife, UFRPE, 2016.

Traits	Relative contribution (%)
Fruit length (cm)	40.71
Fruit width (cm)	35.96
Fruit length/ width ratio	14.08
Number of fruits per plant	5.56
Yield per plant (g)	2.88
Fruit mass (g)	0.81
Total	100

these traits (Table 3).

According to Rohlf (2000), the adjustment of cophenetic correlation coefficient is considered good when values are equal to or higher than (r) 0.70. In this case, the greater the (r) the smaller the distortion of the cluster, presenting a good fit between the matrix and the formed dendrogram (Cruz *et al.*, 2012).

Eggplant genotypes, under high temperatures, showed significant genetic divergence for all evaluated traits. Tocher's optimization methods and the hierarchical UPGMA agreed in 87.50% of genotypes clustering. The traits that the most contributed to divergence were fruit length, fruit width and fruit length/width ratio. The cophenetic correlation coefficient (r) was 0.79. Most genotypes showed genetic similarity with Ciça F1 and Florida Market cultivars.

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