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‘Do-sono’ passion fruit: Ecogeographical prospecting and phenotypic dispersion in transition areas between the *caatinga* and *cerrado* of Brazil

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This study performed eco-geographical prospecting on five populations of wild ‘de-sono’ passion fruit plants in the rural zones of five towns from two distinct agroecological units in the state of Bahia. Ripe fruit from plants from these populations that had fallen to the ground were physicochemically characterized and had their phenotypic variability dispersion among the populations estimated via uni- and multivariate variance and grouping. Among them, the five populations presented univariate and multivariate differentiation. The large majority of multivariate contrasts between the location pairs were shown to be statistically significant Hotelling's T-squared test. The five populations were arranged in a dendogram containing two groups, since a standard of spatial variation between the populations has still not been detected by Mantel Test, although it presents coherence in regards to the agroecological units from which they are allocated. The results are discussed in terms of genetic variability among populations, and the relationships between this diversity, the environmental factors of the ecogeographical units where the plants of the sampled populations were collected, and the implications of the results in terms of selection and use for the genetic improvement of passiflora.

Key words: ‘Do-sono’ passion fruit, native fruits, genetic variability, selection, genetic improvement.

INTRODUCTION

The Passifloraceae family comprises approximately 600 species, about 150 of which are native to Brazil, making our country the principal center of dispersion, and consequently diversity, for this family of species (Oliveira and Ruggiero, 2005; Nascimento, 2006). The cultivation of passiflora stands out in Brazil, considered to be the leading producer of passion fruit (Santos, 2006). Bahia produces about 77 thousand tons of fruit on an area of 7.8 thousand hectares of passiflora, making the state the biggest national producer (Roncatto et al., 2005).

According to Ferreira (1998), more than 50 species of passiflora are primarily cultivated, due to the nutritional

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Table 1. Classification of large geoenvironmental units from the towns in which samples of wild populations of 'do-sono' passion fruit (*P. setacea)* were collected*.*

City	Large Landscape" Unit Chapada Diamantina"		Large Landscape Unit "Superfícies Retrabalhadas		Geoenvironmental Unit in which the collections were taken/samples were collected
	UG C5[1]	UG C7[2]	UG E2[3]	UG E7	
Vitoria da Conquista/VC	Yes	No	Yes	Yes	C5
Piripá/PP	Yes	Yes	No	No	C7
Mortugaba/MO	No	Yes	No	No	C7
Cordeiros/CO	No	Yes	No	No	C7
Condeúba/CD	No	Yes	No	No	C7

Note: [1], [2] and [3] = C5, C7 and E2 = 'Planalto de Vitória da Conquista' and 'Piemontes dos Altos Maciços Centrais and Relevos Associados' 'Maciços and Serras Altas', respectively. [1] The meaning of the abbreviations of the populations is found in Table 2; [2] General average; [3] variation coefficient, in percentage.

qualities of their fruits and pharmaceutical properties of their juices, skins and seeds. Among these is highlighted the 'do-sono' passion fruit (*Passiflora setacea* DC)*,* a plant native to the *cerrado* and *caatinga* biomes in Brazil (Oliveira e Ruggiero; 2005). Its unique fruit, smell and flavors are highly valued in the regions where it occurs, and it is sold in open-air markets of some cities in the Brazilian Northeast (Oliveira, Personal communication, 2010) and the Central-West.

Because they are not commercially grown, fruits of this species are collected from different populations of naturally occurring plants thus explaining the enormous diversity of the physicochemical characteristics of the fruits found for sale in these markets. Species of native plants that are sexually reproduced and have not yet been domesticated, such as the 'do-sono' passion fruit variety, exhibit a lot of heterogeneity in regard to their fruits, both in terms of their physical and chemical characteristics (Cardoso-Silva et al., 2007).

A good survey of functional biodiversity in the scope of vegetable genetic resources, as is the case of characteristics related to the shape and flavor of fruits, requires that the sampling is done to reach the greatest quantity of useful genetic variation in the lowest number of samples possible, keeping in mind the type of phenotypic variability desired. Conceived by Maxted et al. (1995), the methodology of 'ecogeographical prospecting' responds to these issues by taking geographical and ecological aspects into consideration for the purpose of definition of sampling points.

Presently, only one study of the morpho-agronomic dispersion of species of the *Passiflora* spp. genus, based on the principal of 'ecogeographical prospecting' has been done by Araújo (2007), who studied the distribution of the 'do-mato' passion fruit (*Passiflora cincinnata* Mast) in ecogeographical areas of the Brazilian northeast.

This study involves the ecogeographical prospect and study of the dispersion of physicochemical phenotypic variability of fruits via uni- and multivariate variance and grouping analyses of five natural populations of 'de-sono' passion fruit, available in the southwest of Bahia pertaining to geoenvironmental units of the northeast, in order to guide future collection studies focused on the preservation and pre-improvement of the species.

MATERIALS AND METHODS

Ecogeographical collection of the fruits

The fruits were collected from plants from five populations, located in the Bahian municipalities of Vitória da Conquista, Cordeiros, Piripá, Condeúba and Mortugaba, pertaining to two ecogeographical areas in the state of Bahia (Table 1), according to terminology in the Northeast Agroecological Zoning (ZANE) (Silva et al., 2000). Four of these populations of plants are located in GU-C5 and one is in GU-C7.

The choice of these populations and municipalities was due to ease of access and native knowledge about the occurrence of 'do-sono' passion fruit in the region. Local communities make use of the fruit in their diets, and/or make the fruit available for commercialization in markets, for example CEASA in Vitória da Conquista, Bahia.

The number of fruits collected for each population was unequal, due to the variation of availability of the same (population) at the time of collection. The fruits were collected when they were completely ripe, having fallen to the ground in the morning. The particularity of collecting fallen fruit below the dossel of vigorous plants in the process of fruiting, which overlap their vines, impeded counting the number of fruits per plant, or even the number of plants evaluated in each population. Georeferencing data of the collection locations and number of fruits per population are found in Table 2.

After being collected, the fruits were packed in plastic bags, marked and transported for washing and manual de-pulping at the Plant Genetics Laboratory at the Vitória da Conquista campus of the Universidade Estadual do Sudoeste da Bahia [*State University of Southwest Bahia*] (UESB).

Physicochemical characterization of the fruit

The following physical and chemical variables of the ripe fruits were measured: fruit weight (FW) in g; conformity index (CI = LD/TD,

Table 2. Populations of ‘do-sono’ passion fruit (*Passiflora setacea* DC) sampled in five populations, with data on collection location, sample size, geographic coordinates and altitude of collection location.

Population	Location	Total Number of fruits	Number of fruits post-outlier detection	Longitude (West)	Latitude (South)	Altitude (m)
VC	Vitória da Conquista	112	104	42° 19' 49.1"	15° 05' 30.3"	999
MO	Mortugaba	14	14	40° 48' 12.3"	14° 53' 05.4"	1015
CO	Cordeiros	17	17	41° 57' 00.8"	14° 55' 09.7"	704
CD	Condeúba	20	18	41° 58' 38.5"	14° 52' 14.7"	739
PP	Piripá	25	22	42° 05' 18.4"	14° 40' 28.0"	820

where, LD = lateral diameter and TD = transversal diameter), pulp weight (PW) in g; pulp yield [PY (%) = FW - SW/FW x 100, where SW = shell weight); shell thickness (ST) in mm; pH; total soluble solid level (TSS); citric acid percent (CA%) and proportion of soluble solids to acidity (TSS/CA%). The measurement of the variables ‘proportion of total soluble solids’ and citric acid percent were carried out according to Cardoso-Silva et al. (2007). The weight measurements in grams were obtained using a digital scale, and the dimensions in millimeters were obtained with a digital caliper.

Uni- and multivariate analysis

Prior to performing uni- and multivariate statistical procedures, the existence of extreme values was determined based on the Outliers-base function in the deviances (α = 0.05) BioEstat 4.0 (Ayres et al., 2005). The remaining quantity of fruits in the population after removal of extremes is found in Table 2.

The measurements of each physicochemical variables of fruit were submitted to unbalanced (α = 0.01) variance analysis (ANAVA) considering an entirely random statistical delineation with five treatments (populations) and fruits per plant used as repetitions. The ANAVA was carried out using BioEstat 4.0 software (Ayres et al., 2005).

The set of new measurements of the variables was submitted to unbalanced multivariate analysis of variance (Manova; α = 0.01), now considered a ‘source of variation’ the (*i*) two distinct ecogeographical regions (Pillai’s trace, α = 0.01), or (ii) the five wild populations, independent of ecogeographical origin (Pillai’s trace, α = 0.01).

The significance of the contrasts between location pairs was obtained via a Hotelling T-squared test (α = 0.01) (Kramer, 1972), which expressed the geographic distance between the populations. The MANOVA was carried out using the MINITAB^{TM} (Minitab Inc., 1999) software.

Grouping and Mantel analyses

The variables were then submitted to grouping analysis (UPGMA) through calculation of the average euclidean distance between the populations, followed by adjustment between the distance matrix and the dendogram, employing the cophenetic correlation coefficient (*r*).

Finally, analysis of the spatial variation standard was done in a multivariate context via Mantel analysis (1967), with the geographic distance matrix and the average euclidean distance being used to measure dissimilarity between the populations, with 1000 simulations. Both the grouping analysis used and the Mantel test were performed using the Genes program (Cruz, 2001).

RESULTS AND DISCUSSION

ANAVA has identified that the five plant populations exhibit fruits that are statistically different in regard to physicochemical characteristics of fruit studied (p <0.0001), which denotes differential performance between the populations, and relates the existence of genetic variability sufficient to indicate priority areas for the purpose of collecting and selecting of the best populations of ‘do-sono’ passion fruit. The average values of the characteristics are presented in Table 3.

The morpho-agronomic descriptors were used to determine the genetic variability of the species obtained values of the variation coefficient (VC, %), which varied from 5.5 to 31.8; showing that the variables used present different levels of heterogeneity.

Even in one species, in each of the studied areas, the plants are subject to fluctuations of temperature, rainfall levels and other variants that may influence certain aspects of its genetic composition. That is, the mean may be adequate to express determined characteristics that do not appear in another location (Botezelli et al., 2000), it is what may be reported in the results obtained in the five studied populations.

Analyses of the fruits by collection area identified that the populations from Vitória da Conquista and Condeúba stand out for average values of physicochemical characteristics of fruit being adequate for industry, principally in regard to pulp yield and total soluble solids, and the areas are indicated as adequate for prospecting superior genotypes that may compos germplasm banks for pre-improvement studies and conservation of the species.

Significant multivariate genetic divergences have detected between the two distinct ecogeographical regions (Pillai’s trace, *p value* < 0.001). That is, the geoenvironmental units C5 and C7 show natural populations of ‘do-sono’ passion fruit that genetically diverge among themselves, corresponding to the set of physicochemical characteristics of fruit evaluated.

Each of the five wild populations were tested in MANOVA independent of ecogeographical origin. The *p value* surveyed by the statistic from the Pillai’s trace test (< 0.001) corroborates that these genetically diverge with

Table 3. Results obtained from the physicochemical analysis of five populations of ‘do-sono’ passion fruit.

Population[1]	Fruit weight (FW)	Conformity index (CI)	Pulp weight (PW)	Pulp yield (PY)	Shell thickness (ST)	pH	Total soluble solid level	Citric Acid (%)	Proportion of soluble solids to acidity (TSS/CA)
VC	48.48	1.27	27.18	55.51	3.29	3.21	15.56	2.04	7.92
MO	33.60	1.14	15.18	43.99	2.38	3.40	15.22	2.35	6.49
CO	40.63	1.13	20.84	49.73	4.92	2.94	13.71	2.71	5.60
CD	42.37	1.20	20.93	48.85	3.47	3.33	11.90	3.46	4.10
PP	55.41	1.10	27.42	50.40	5.37	3.02	14.39	2.54	6.52
QMT	0.0014	0.20	663.39	631.12	34.79	0.69	65.19	9.61	76.57
QMR	234.74	0.03	92.20	83.65	1.56	0.07	6.83	0.83	6.84
X[2]	44.10	1.17	22.31	49.70	3.89	3.18	14.16	2.62	6.13
CV(%)[3]	18.70	5.5	22.95	8.28	31.77	6.11	10.28	20.19	22.93

[1] The meaning of the abbreviations of the populations is found in Table 2; [2] General average; [3] variation coefficient, in percentage.

Table 4. Values from the Hotelling T^2 multivariate and their respective *p-value*, among five wild populations of ‘do-sono’ passion fruit (*P. setacea* DC) belonging to the geoenvironmental units C5 and C7.

Population	VC	MO	CO	CD	PP
VC[1]	-	63.84**	122.85**	299.7**	86.62**
MO			73.92**	65.55**	134.46**
CO				83.98**	21.3*
CD					134.54**
PP					-

Note.: [1] The meaning of the abbreviations of the populations is found in Table 2; ** *p-value* < 0.0001; * *p-value* = 0.043

each other, with at least one contrast pair existing among the populations with multivariate differences. The Hotelling T^2 test (α = 0.01) was used to test the two-to-two contrasts; values surveyed from the test, and their degree of corresponding statistical significance, are found in Table 4.

With the exception of the multivariate contrast between the pair of populations of PP and CO (*p value* = 0.043), a significant multivariate difference was detected (*p value* < 0.0001) for all of the other contrasts. It is worth noting that the geographic distance between populations PP and CO was the lowest among all of the contrast pairs (6 km) (Table 5). The Hotelling T^2 statistic corroborates the genetic differences detected by the individual ANAVAs. With the existence of differences among the vectors of average for the populations proven with the Hotelling T^2 test, the average euclidean distances of the five populations were calculated (Table 5).

In regard to genetic differences, populations CO and PP showed the lowest euclidean distance values (0.82), and the highest values were found for populations VC and CD (1.74). The non-contrast between CO and PP may be explained by the fact that both of these populations are

Table 5. Distance matrix among the five populations of 'do-sono' passion fruit plant (*P. setacea* DC), based on the measurement of nine physicochemical characteristics of fruit.

Population[1]	VC	MO	CO	CD
VC	-			
MO	1.61	-		
CO	1.44	1.31	-	
CD	1.74	1.41	1.12	-
PP	1.28	1.68	0.82	1.49

[1]The meaning of the abbreviations of the populations is found in Table 2.

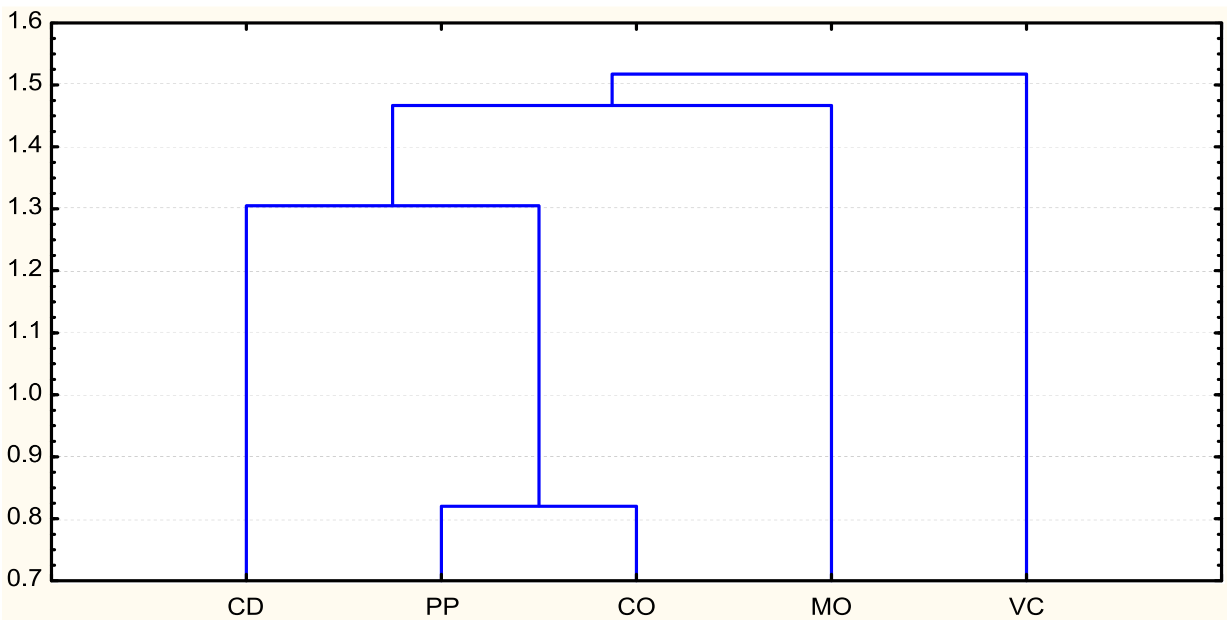

Figure 1. Dendogram of dissimilarity based on the average euclidean distance from the beginning of nine physical and chemical characteristics of fruit from five wild populations of 'do-sono' passion fruit (P. setacea DC), occurring in two geoenvironmental units in Bahia. The cophenetic correlation coefficient for the dendogram was 0.786.

found in the same geoenvironmental unit (GU-7), which shows more homogenous edaphoclimatic conditions, as well as due to the close physical proximity of these populations. The plants from CO and PP are more closely related as a result of the occasional gene flow between these two populations.

The greater genetic divergence found between populations VC and CD may be explained by ecogeographic origin. The two populations are located in distinct GUs [C5 (the population of Vitória da Conquista) and C7 (population of Condeúba)]. Considering this approach, note that the greatest genetic differences were related to the different geoenvironmental units, which can probably be explained by the greater geographic variability found, which may be associated with differences in soil type, elevation and precipitation. Based on these distances the hierarchical grouping was generated using the UPGMA method (Figure 1), which validates the results of the multivariate Hotelling T^2 test.

The formation of two groups can be noted. In the first group, the population of Vitória da Conquista is ordered in an isolated manner. It should be emphasized that this population is the only one among the five studied in this work pertaining to GU-C5. The second group was made up of populations from Mortugaba and a subgroup containing the populations from Condeúba, Cordeiros and Piripá (Figure 1). All four of these populations are in the UG-C7 (Figure 1).

The standards of similarity between the populations surveyed by the UPGMA may be analytically understood through the use of the simple Mantel test. The correlation coefficient between the genetic distance matrix and the geographic distance showed a positive yet insignificant value (r = 0.35; p > 0.05). The structure in the space presented should not have a direct influence on the geographic distribution on these characteristics. The phenotypes of the populations may be influenced by other factors such as soil type, elevation, pluviometer, etc.; they are found independent of spatial distribution and genetic divergence, which indicates a possible adaptive process or response by a reaction standard to the environmental variations in these populations (Morales, 2000).

Finally, the use of the strategy of ecogeographical prospecting (Maxted et al., 1995) is an important tool for research of functional biodiversity prospecting of native passion fruit, similar to this study, in which genotypic variants presenting different magnitudes of physicochemical characteristics of fruit may be ecogeographically located and defined, for later use under the scope of pre-improvement conservation.

Conclusion

For the majority of physical and chemical characteristics of the fruit, there was high phenotypic variability in the populations of ‘do-sono’ passion fruit studied; indicating that genetic gain through selection is possible.

The locations of Vitória da Conquista and Piripá are recommended for prospecting plants with fruit adequate for industry, principally due to pulp yield and proportion of total soluble solids.

The phenotypes of the populations of ‘do-sono’ passion fruit are probably influenced by the ecogeographic conditions where they are found, independent of distribution in space and their genetic differentiation, since the Mantel test was not significant, suggesting that an adaptive process exists that is playing on the environmental variations in these populations.

The adoption of consumption at the regional level endows the ‘do-sono’ passion fruit plant, which occurs naturally in the biomes of the Brazilian *caatinga* and *cerrado*, with the potential for future genetic domestication, with the goal of synthesis and selection of regional commercial varieties.

Conflict of Interests

The author(s) have not declared any conflict of interests.

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