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REPEATABILITY OF FRUIT QUALITY TRAITSOF CACTUS PEAR[1]

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ABSTRACT - Repeatability analysis has been used to study traits in several crops, assisting in the definition of the minimum number needed to evaluate genotypes more efficiently and with less time and resource consumption. So far, however, no repeatability studies on cactus pear have been found in the literature. The objective of this study was to determine the coefficient of repeatability for cactus pear fruits traits and the minimum number of evaluations (fruit) that can provide acceptable accuracy for the prediction of the true value. The experiment was conducted at the Federal Institute of Bahia/Campus Guanambi, with 150 fruits collected from three municipalities in the state of Bahia. The coefficients of repeatability were estimated by the methods of analysis of variance, principal components based on the covariance (PCCV) and correlation (PCC) matrices, and structural analysis based on the correlation matrix (SA). The analysis of variance showed that, except for fruit diameter, the effect of the production site (municipality) was significant for all traits evaluated. The PCCV method was proven the most suitable for studying the repeatability of quality traits of cactus pear fruits. Seven fruits were required to determine, with 90% confidence, the traits length, diameter, fruit firmness, skin thickness, number of seeds, fruit mass, bark mass, pulp mass, pH, titratable acidity, soluble solids, SS/AT ratio, and pulp yield.
Index terms: *Opuntia ficus-indica*, sample size, components of variance, phenotypic covariance.

REPETIBILIDADE PARA CARACTERES DE QUALIDADE DE FRUTOS DE FIGUEIRA DA ÍNDIA

RESUMO – A análise de repetibilidade tem sido utilizada para estudo de caracteres de diversas culturas, auxiliando na definição do número mínimo necessário para avaliações de genótipos com maior eficiência e menor consumo de tempo e recursos. Todavia, para a figueira da índia não foi encontrada na literatura pesquisas dessa natureza. Assim, este estudo teve como objetivo determinar o coeficiente de repetibilidade para características do fruto de figueira da índia e o número mínimo de avaliações (fruto) capaz de proporcionar confiabilidade aceitáveis para a predição do valor real. O trabalho foi desenvolvido no Instituto Federal Baiano/*Campus* Guanambi, analisando-se 150 frutos, provenientes de três municípios do estado da Bahia. Os coeficientes de repetibilidade foram estimados pelos métodos da análise de variância, componentes principais com base na matriz de covariância (CPCV) e de correlações (CPC), e análise estrutural com base na matriz de correlações (AE). A análise de variância mostrou que, com exceção do diâmetro dos frutos, o efeito do local de produção (município) foi significativo para todas as características avaliadas. O método dos CPCV demonstrou ser o mais adequado para o estudo da repetibilidade para características de qualidade dos frutos da figueira da índia. São necessários 7 frutos, para determinar, com 90% de confiança, os caracteres comprimento, diâmetro, firmeza do fruto, espessura da casca, número de sementes, massa do fruto, massa da casca, massa da polpa, pH, acidez titulável, sólidos solúveis, relação SS/AT e rendimento de polpa em figueira da índia.
Termos para indexação: *Opuntia ficus-indica,* tamanho de amostra, componentes de variância, covariância fenotípica.

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INTRODUCTION

Cactus pear (*Opuntia ficus-indica* (L.) Mill.) is a species of cactus originated from Mexico and cultivated for fruit production in many parts of the world, mainly under rainfed conditions, with important commercial plantations in Italy, Israel, and Mexico (MANICA, 2002).

Cactus pear has attracted increasing attention in both domestic and export markets for its nutritional health promotion benefits due to its contents of fiber, soluble carbohydrates, calcium, magnesium, and vitamins (mainly A and C); as well as the possibility of exploiting medicinal properties, with several biological activities, including antioxidant and anti-inflammatory properties (GALATI et al., 2003; ZHONG et al., 2010; YAHIA and JACOBO, 2011).

In Brazil, the plant is named forage palm and is mostly cultivated as animal fodder, with extensive cultivated areas mainly in the northeastern semi-arid region (PINHEIRO et al., 2014), though the fruits have been exploited through extractivism. Thus, cactus pear production has a high potential for price and income aggregation, which may lead to improvements in the quality of life for the Brazilian semiarid population. However, the varieties of this crop available were selected for animal feeding, pointing out the need to search for genetic materials that produce quality fruits and the characterization of the genetic variability to initiate breeding works.

In the stages of characterization of germplasm bank accessions or even to perform characterization of fruits, a recurrent question among researchers is about the number of fruits that make a sample representative of the population to be studied. Knowing the number of sample units to be collected in prospecting expeditions is crucial. This information is important for scheduling collection expeditions, in order to optimize the time and area required for collecting and transporting genetic material that can be incorporated into breeding programs (Manfio et al., 2011).

According to Cardoso (2006), breeding programs tend to reduce or eliminate the time spent with evaluating repetitions more than it is necessary, as well as tend to avoid the evaluation of a small number of repetitions that would incur probable estimation errors, leading to failure in the identification of superior genotypes. Therefore, knowing the coefficient of repeatability allows the plant breeder to carried out the evaluation phase efficiently, optimizing the time, financial resources, and labor.

Repeatability expresses the maximum value that the heritability can reach, because it expresses the proportion of the phenotypic variance, which is attributed to the genetic differences confounded with the permanent effects that affect the cultivar or progeny. Thus, repeatability, like heritability, is an indispensable tool for guiding improvement work. In this sense, high estimates of the repeatability of a trait indicate that it is possible to predict the true value of the individual with a relatively small number of measurements (Cruz et al., 2012).

The repeatability analysis has been applied to several species, including bacabi (*Oenocarpus mapora* Karsten) (OLIVEIRA and MOURA, 2010), araça (*Psidium cattleyanum)* and pitanga (*Eugenia uniflora)* (DANNER et al., 2010); passionfruit (NEVES et al., 2010), macaw palm (*Acrocomia aculeata)* (MANFIO et al., 2011), peach (BRUNA et al., 2012); soybean (MATSUO et al., 2012), peach palm (*Bactris gasipaes)* (BERGO et al., 2013), and sweet orange (NEGREIROS et al., 2014). However, no repeatability studies on cactus pear fig have been found in the literature so far.

Thus, the objective of this work was to determine the repeatability coefficient of of cactus pear fruit traits and the minimum number of evaluations (sample units) capable of providing confidence to predictors of the true value.

MATERIAL AND METHODS

The study was conducted at the Instituto Federal Baiano - Campus Guanambi, located in the municipality of Guanambi/BA, using for the analyses cactus pear fruits of cv. Gigante from crops intended for animal feed, without cultural managements of irrigation, fertilization, and control of pests and diseases. Fruits were harvested from small farms located in three municipalities in the semi-arid region of the state of Bahia: Guanambi located at 14º13’24’’ south latitude and 42º46’53’’ west longitude, 530 m altitude; Pindaí located at 14º29’33’’ south latitude and 42º41’14’’ west longitude, 610 m altitude; and Riacho de Santana located at 13º36’33’’ south latitude and 42º56’20’’ west longitude, 627 m altitude.

Fifty fruits were collected randomly from plants of the same crop of each municipality, at the same maturation stage, for analyses of physical and physico-chemical characteristics. The following physical characteristics were measured: longitudinal length and diameter by direct measurement using a digital caliper and expressed as centimeters; mass of fruit, skin, and pulp by individual weighing on a digital analytical scale and expressed as grams; firmness of the whole fruit with skin measured individually at two distinct points of the equatorial region on the intact fruit uaing a digital dynamometer and expressed as Newtons (N); number of seeds per fruit by counting in each fruit, and pulp yield by dividing pulp mass by fruit mass and multiplying by 100, and expressed as percentage.

The physico-chemical characteristics were: pH by direct reading using a bench potentiometer with glass membrane electrode calibrated with solutions of pH 4.0 and 7.0; titratable acidity by titration with 0.1N sodium hydroxide using 1% phenolphthalein as indicator and expressed as % of citric acid; soluble solids by direct reading in a digital refractometer and expressed as ºBrix; and the SS/AT ratio obtained by dividing soluble solids by titratable acidity and expressed as absolute value, to two decimal places. All physico-chemical analyses were performed following the methodologies of the Analysis Manual of the Instituto Adolfo Lutz (IAL, 2008).

Estimates of repeatability coefficients were obtained by the methods of analysis of variance, principal components based on the covariance (PCCV) and correlation (PCC) matrices, and structural analysis based on the correlation matrix (SA).

The estimates of the repeatability coefficients, number of measurements required to predict the true value of the individuals, and the genotypic determination were based on the statistical models described by Cruz et al. (2012) and obtained by the repeatability procedure of the GENES program (CRUZ, 2013).

RESULTS AND DISCUSSION

The analysis of variance showed that, except for fruit diameter, the effect of the production site (municipality) was significant for all traits evaluated (Table 1), indicating the existence of environmental variability among the sites.

The repeatability coefficients ($\hat{r}$) estimated by the methods Anova and Structural Analysis (AE) provided the lowest values, independent of the trait analyzed, thus, a larger number of fruits are needed for the evaluation in order to provide confidence to the estimates obtained through these methodologies (Table 2).

The univariate method (Anova) provides the lowest repeatability coefficients because it does not allow us to isolate the component of genotypic variance used to estimate the repeatability, which is part of the variance of the environment (COSTA, 2003) and when is incorporated into the estimate, it increases the experimental error and may underestimate the repeatability (NEGREIROS et al., 2014). This fact is evident, due to the significant difference found among the production sites by the analysis of variance.

In this case, the repeatability coefficient is more efficiently estimated by the principal components of the covariance matrix, which takes into account the cyclic behavior of the trait (DANNER et al., 2010), because the alternating effect is included into the experimental error (Manfio et al., 2011). In similar studies carried out with hybrids of African Oil Palm (*Elaeis guineensis* Jacq.) and Brazilian Oil Palm (*Elaeis oleifera* (H.B.K.) Cortés) (Chia et al., 2009), araça (*Psidium cattleyanum)* and pitanga (*Eugenia uniflora)* (DANNER et al., 2010); peach palm (*Bactris gasipaes)* (BERGO et al., 2013), sweet orange (NEGREIROS et al., 2014), and kale (*Brassica oleracea*) (AZEVEDO et al., 2016), the estimates obtained by the Anova method were always lower than those obtained by the multivariate analysis because, according to Cruz et al. (2012), the Anova method does not remove the environmental effect of alternating production in different years, underestimating the repeatability coefficient.

Fruit diameter and skin thickness showed the greatest variations for the repeatability coefficient among the different methods, with repeatability estimates ranging from 0.002 to 0.57 (ANOVA and PCCV) for diameter and 0.04 to 0.59 (ANOVA and PCCV) for thickness. However, the lowest repeatability variations were found for titratable acidity and soluble solids, with values ranging from 0.95 to 0.99 (ANOVA and PCCV), which indicates a certain regularity in the repetition of the trait between the evaluations. Bergo et al. (2013) reported a similar behavior for morphological traits of peach palm, and pointed to PC as the method that allows the highest coefficient of repeatability, considering as confident $\hat{r}$ values above 0.4, but with a greater number of measurements over the growing cycles. Neves et al. (2010) also suggested the use of a greater number of measurements for passion fruit; however, Manfio et al. (2011) and Bruna et al. (2012), evaluating the perennial plants macaw palm and peach, respectively, considered values above 0.6 as high coefficient of repeatability.

Despite the variation in the $\hat{r}$ estimates, the high values obtained by the multivariate methods, especially PCCV, indicate regularity in the repetition of clone performance over the evaluations (Cruz et al., 2012).

Using the PCCV method, the minimum number of fruits required to predict the true value of the clone, with 95% determination coefficient for fruit length (2), fruit diameter (14), fruit mass (3), pulp mass (3), skin mass (15), pulp yield (8), skin thickness (13), number of seeds (11), firmness (7), soluble solids (1), titratable acidity (1), soluble solids/titratable acidity ratio (3), and pH (6) will be 15 fruits, as it was established for the trait skin mass, since the estimates of the other traits are included in this value, and fruit quantities over 11 for the traits fruit diameter, skin mass, skin thickness, and number of seeds are due to their larger effect of the interaction genotype x environment (AZEVEDO et al., 2016).

It is noteworthy that for many traits evaluated, the number of fruits/measurements required is just one fruit, which, according to Cargnin (2016), is due to high heritability estimates, that is, traits that are not much influenced by the environment, and because the genetic material derives from clones.

Chia et al. (2009) argues that, within acceptable levels of precision, one should reduce the number of evaluations to save time and resources. A number of studies have considered 80-90% as good accuracy (CHIA et al., 2009; DANNER et al., 2010; MANFIO et al., 2011; MATSUO et al., 2012).

This study showed that, with a determination coefficient of 90%, it is possible to reduce the number of evaluations for fruit length (1), fruit diameter (6), fruit mass (2), pulp mass (1), skin mass (7), pulp yield (4), skin thickness (6), number of seeds (5), firmness (3), soluble solids (1), titratable acidity (1), soluble solids/titratable acidity ratio (1), and pH (3) to 7 fruits. Thus, evaluations performed on 7 fruits allow us to obtain accurate estimates for the traits assessed in this study, with good confidence and reduction in operational expenses.

TABLE 1 - Analysis of variance for the traits analyzed in cactus pear fruits collected in three municipalities of the State of Bahia.

Trait				
Length	39.73	0.42	8.64	7.50
Diameter	0.217^{ns}	0.19	5.81	7.69
Fruit mass	30446.90*	388.92	171.59	11.49
Pulp mass	15029.44*	199.75	91.75	15.40
Skin mass	4244.56*	153.38	75.83	16.33
Pulp yield	1094.84*	70.78	53.64	15.68
Skin thickness	2.95*	0.64	2.00	40.0
N° Seeds	11917.04*	338.96	291.36	6.31
Firmness	3.39*	0.67	27.06	3.02
Soluble solids	645.19*	0.53	11.70	6.27
Titratable acidity	14.49*	0.01	0.50	13.39
AT/SS ratio	36220.62*	171.98	42.97	30.51
pH	11.09*	0.12	5.58	6.36

* mean squared for municipalities evaluated, MSE – mean square of error and CV (%) – coefficient of variation. ^{ns}Non significant, * significant at1% probability by the F test.

TABLE 2 - Estimation of the coefficients of repeatability ($\hat{r}$),determination (R^2), and minimum number of fruits/measurements by the methods of analysis of variance (ANOVA), principal components based on correlation matrix (PCC) (PCCV), and structural analysis based on the correlation matrix (SA) of cactus pear fruits collected in three municipalities of the State of Bahia

Trait	Value obtained from 50 measurements			$R^{2(1)}$	ANOVA η_0	PCCV η_0	PCC η_0	SA η_0
		$\hat{r}$	R^2 (%)	0.80	2.14 (2)	0.44 (1)	0.77 (1)	2.34 (2)
	ANOVA	0.65	98.94	0.85	3.03 (3)	0.63 (1)	1.10 (1)	3.31 (3)
Fruit length	PCCV	0.89	99.78	0.90	4.82 (5)	1.00 (1)	1.74 (2)	5.26 (5)
	PCC	0.83	99.61	0.95	10.17 (10)	2.11 (2)	3.68 (4)	11.11 (11)
	SA	0.63	98.84	0.99	53.03 (53)	11.01 (11)	19.21 (19)	57.93 (58)
		$\hat{r}$	R^2 (%)	0.80	2230.28 (2230)	2.90 (3)	3.04 (4)	258.05 (258)
	ANOVA	0.002	8.22	0.85	3159.57 (3160)	4.12 (4)	4.30 (4)	365.57 (365)
Fruit diameter	PCCV	0.57	98.56	0.90	5018.14 (5018)	6.54 (6)	6.843 (7)	580.61 (580)
	PCC	0.56	98.5	0.95	10593.85 (10594)	13.81 (14)	14.44 (14)	1225.7 (1225)
	SA	0.01	43.66	0.99	55199.55 (55200)	71.99 (72)	75.27 (75)	6386.75 (6386)
		$\hat{r}$	R^2 (%)	0.80	2.58 (2)	0.71 (1)	1.26 (1)	2.89 (3)
	ANOVA	0.60	98.72	0.85	3.66 (3)	1.00 (1)	1.79 (2)	4.09 (4)
Fruit mass	PCCV	0.84	99.64	0.90	5.82 (6)	1.59 (2)	2.85 (3)	6.51 (6)
	PCC	0.75	99.37	0.95	12.29 (12)	3.37 (3)	6.02 (6)	13.74 (14)
	SA	0.58	98.57	0.99	64.04 (64)	17.57 (17)	31. 37 (31)	71.61 (72)
Pulp mass		$\hat{r}$	R^2 (%)	0.80	2.69 (2)	0.58 (1)	1.05 (1)	2.45 (2)
	ANOVA	0.59	98.67	0.85	3.81 (4)	0.82 (1)	1.49 (1)	3.47 (3)
	PCCV	0.87	99.70	0.90	6.06 (6)	1.31 (1)	2.37 (2)	5.51 (5)
	PCC	0.79	99.47	0.95	12.79 (13)	2.77 (3)	5.01 (5)	11.65 (11)
	SA	0.61	98.78	0.99	66.67 (67)	14.47 (14)	26.10 (26)	60.71 (61)
Skin mass		$\hat{r}$	R^2 (%)	0.80	7.498 (7)	3.26 (3)	3.98 (4)	8.11 (8)
	ANOVA	0.34	96.18	0.85	10.62 (11)	4.62 (5)	5.64 (6)	11.50 (11)
	PCCV	0.55	98.39	0.90	16.87 (17)	7.34 (7)	8.95 (9)	18.26 (18)
	PCC	0.50	98.04	0.95	37.67 (38)	15.51 (15)	18.91 (19)	38.56 (38)
	SA	0.33	96.09	0.99	196.31 (196)	80.83 (80)	98.54 (98)	200.92 (201)
Pulp yield		$\hat{r}$	R^2 (%)	0.80	13.82 (14)	1.65 (2)	2.38 (2)	9.14 (9)
	ANOVA	0.224	93.53	0.85	19.58 (20)	2.34 (2)	3.37 (3)	12.95 (13)
	PCCV	0.707	99.17	0.90	31.10 (31)	3.72 (4)	5.35 (5)	20.57 (20)
	PCC	0.627	98.82	0.95	65.66 (66)	7.87 (8)	11.30 (11)	43.43 (43)
	SA	0.304	95.62	0.99	342.16 (342)	41.02 (41)	58.89 (59)	226.32 (226)
Skin thickness		$\hat{r}$	R^2 (%)	0.80	55.17 (55)	2.75 (3)	2.75 (3)	96.0 (96)
	ANOVA	0.06	78.37	0.85	78.16 (78)	3.90 (4)	3.90 (4)	136.0 (136)
	PCCV	0.59	98.63	0.90	124.13 (124)	6.20 (6)	6.20 (6)	216.0 (216)
	PCC	0.59	98.63	0.95	262.06 (262)	13.10 (13)	13.10 (13)	456.0 (456)
	SA	0.040	67.56	0.99	1365.51 (1365)	68.27 (68)	68.27 (68)	2376.0 (2376)

N° Seeds		$\hat{r}$	R^2 (%)	0.80	5.855 (6)	2.43 (2)	2.25 (2)	3.77 (4)
	ANOVA	0.49	97.15	0.85	8.29 (8)	3.44 (3)	3.19 (3)	5.34 (5)
	PCCV	0.62	98.79	0.90	13.17 (13)	5.47 (5)	5.07 (5)	8.48 (8)
	PCC	0.63	98.88	0.95	27.81 (28)	11.55 (11)	10.72 (11)	17.91 (18)
	SA	0.514	98.14	0.99	144.92 (145)	60.22 (60)	55.87 (56)	93.36 (93)
Firmness		$\hat{r}$	R^2 (%)	0.80	49.36 (49)	1.39 (1)	1.86 (2)	660.94 (661)
	ANOVA	0.07	80.20	0.85	69.93 (70)	1.97 (2)	2.64 (3)	936.33 (936)
	PCCV	0.74	99.30	0.90	111.07 (111)	3.14 (3)	4.20 (4)	1487.11 (1487)
	PCC	0.68	99.07	0.95	234.49 (234)	6.63 (7)	8.87 (9)	3139.47 (3139)
	SA	0.07	23.23	0.99	1221.83 (1222)	34.56 (34)	46.24 (46)	16358.30 (16358)
Soluble solids		$\hat{r}$	R^2 (%)	0.80	0.16 (1)	0.08 (1)	0.09 (1)	0.09 (1)
	ANOVA	0.95	99.91	0.85	0.23 (1)	0.12 (1)	0.12 (1)	0.12 (1)
	PCCV	0.97	99.96	0.90	0.37 (1)	0.19 (1)	0.20 (1)	0.20 (1)
	PCC	0.97	99.95	0.95	0.79 (1)	0.43 (1)	0.43 (1)	0.43 (1)
	SA	0.97	99.95	0.99	4.13 (4)	2.11 (2)	2.24 (2)	2.25 (2)
Titratable acidity		$\hat{r}$	R^2 (%)	0.80	0.06 (1)	0.01 (1)	0.01 (1)	0.01 (1)
	ANOVA	0.98	99.96	0.85	0.09 (1)	0.01 (1)	0.02 (1)	0.02 (1)
	PCCV	0.99	99.99	0.90	0.14 (1)	0.04 (1)	0.03 (1)	0.03 (1)
	PCC	0.99	99.99	0.95	0.30 (1)	0.006 (1)	0.06 (1)	0.07 (1)
	SA	0.99	99.99	0.99	1.57 (1)	0.33 (1)	0.36 (1)	0.36 (1)
Titratable acidity/ Soluble solids ratio		$\hat{r}$	R^2 (%)	0.80	0.95 (1)	0.64 (1)	0.63 (1)	0.66 (1)
	ANOVA	0.80	99.52	0.85	1.35 (1)	0.91 (1)	0.89 (1)	0.94 (1)
	PCCV	0.86	99.67	0.90	1.45 (2)	1.45 (1)	1.42 (1)	1.49 (1)
	PCC	0.86	99.68	0.95	4.53 (4)	3.07 (3)	3.01 (3)	3.15 (3)
	SA	0.85	99.66	0.99	23.61 (23)	16.00 (16)	15.68 (16)	16.42 (16)
pH		$\hat{r}$	R^2 (%)	0.80	2.30 (2)	1.21 (1)	1.54 (1)	1.99 (2)
	ANOVA	0.63	98.85	0.85	3.26 (3)	1.71 (1)	2.18 (2)	2.82 (3)
	PCCV	0.76	99.39	0.90	5.19 (5)	2.72 (3)	3.47 (3)	4.48 (4)
	PCC	0.72	99.23	0.95	10.95 (11)	5.75 (6)	7.33 (7)	9.47 (9)
	SA	0.66	99.01	0.99	57.09 (59)	29.98 (30)	38.23 (38)	49.35 (49)

*Coefficient of determination of the method Number calculated (approximate number).

CONCLUSIONS

There is significant environmental variability among the three municipalities for the quality traits evaluated on cactus pear fruits.

The coefficient of repeatability is efficiently estimated by the principal components method based on the covariance matrix.

Seven fruits are sufficient to evaluate the physical and physico-chemical traits of cactus pear with determination coefficient of 90%.

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