ISSN 0100-2945 http://dx.doi.org/10.1590/0100-29452017003

GENETIC AND MORPHOAGRONOMIC DIVERSITY OF *Passiflora* spp. BASED ON QUANTITATIVE MEASUREMENTS OF FLOWERS AND FRUITS[1]

JAMILE DA SILVA OLIVEIRA[2], FÁBIO GELAPE FALEIRO[3], NILTON TADEU VILELA JUNQUEIRA[3], MARCELO LIBINDO VIANA[4]

ABSTRACT- The aim of this study was to characterize *Passiflora* spp. accessions and its genetic diversity based on quantitative morphological descriptors of flowers and fruits. The study was conducted at Embrapa Cerrados, Planaltina-DF. Fifteen *Passiflora* spp. accessions were characterized using 14 quantitative morphological descriptors. Genetic distances among accessions were estimated based on Mahalanobis' generalized distance. Cluster analysis via dendrogram and graphic dispersion was analyzed. The relative contribution of characters for accession divergence was also calculated. The morphoagronomic characterization based on quantitative descriptors of flowers and fruits contributed to the differentiation of *Passiflora* spp. accessions, serving as an important tool for variability quantification. This information is useful to perform *Passiflora* spp. characterization and genetic diversity studies.
Index terms: multivariate analysis, Passifloraceae, genetic resources.

DIVERSIDADE GENÉTICA E MORFOAGRONÔMICA DE *Passiflora* spp. BASEADA EM VARIÁVEIS QUANTITATIVAS DAS FLORES E FRUTOS

RESUMO- Objetivou-se caracterizar acessos de *Passiflora* spp. e sua diversidade genética baseada em descritores morfoagronômicos quantitativos de flores e frutos. O estudo foi realizado na EMBRAPA Cerrados, Planaltina-DF. Foram caracterizados 15 acessos de *Passiflora* spp. utilizando 14 descritores morfoagronômicos quantitativos. Distâncias genéticas entre os acessos foram estimadas com base na Distância de Mahalanobis. Análise de agrupamento via dendrograma e dispersão gráfica via coordenadas principais foram analisadas. Foi calculada, também, a contribuição relativa dos caracteres para divergência genética dos acessos analisados. A caracterização morfoagronômica baseada em descritores quantitativos de flores e frutos contribuiu para a diferenciação fenotípica entre os acessos *Passiflora* spp., servindo como importante instrumento para quantificar a variabilidade. Essa caracterização é importante para estudos mais completos de caracterização e diversidade genética do gênero *Passiflora.*
Termos para indexação: análise multivariada, Passifloraceae, recursos genéticos.

[1](Paper 208-15). Received September 02, 2015. Accepted June, 2016.
[2] Agronomist, D.Sc. Student in Agronomy, Universidade de Brasília, Campus Universitário Darcy Ribeiro, Brasília, DF, CEP 70910-900. E-mail: jamile.oliveira54@gmail.com.
[3] Agronomists, Researchers at Embrapa Cerrados, BR 020 Km 18, Planaltina, DF, Brazil, CEP 73310-970, CP: 08223. E-mails: fabio.faleiro@embrapa.br, nilton.junqueira@embrapa.br
[4] Agronomist, trainee at Embrapa Cerrados, BR 020 Km 18, Planaltina, DF, Brasil, CEP 73310-970, CP: 08223. E-mail: faz.nsa@gmail.com

INTRODUCTION

The use of genetic resources has viability based on collection, introduction, conservation and exchange of germplasm accessions, as well as on their characterization and evaluation, which allows knowing the qualities and potentialities of the material (FALEIRO et al., 2012). In a survey on demands for passion fruit research, Faleiro et al. (2006) indicated the characterization, domestication and use of new species as a priority. An important step of characterization and evaluation processes is the elaboration of descriptors, which can be morphological, agronomic and molecular.

For the development of a breeding program, among other aspects, it is important to characterize accessions in order to obtain basic information about genotypes for use in crosses (FALEIRO et al., 2008). The verification and quantification of intra and interspecific variability is of utmost importance, since it allows genetic resources to be more efficiently used by breeders.

Morphoagronomic descriptors play a fundamental role in the characterization and selection of plants, being decisive in the choice of genotypes along recombination cycles and also in the selection of genotypes for use as new parents. The morphoagronomic characterization for genetic variability studies has been made based on features that are easy to detect and measure and suffer little environmental influence.

Negreiros et al. (2008) characterized yellow passion fruits from half-sibling progenies and observed that some fruits presented desirable characteristics both for the fresh market and for the industry, as well as superiority of some genotypes for future breeding programs.

To characterize *P. edulis* genotypes, Castro et al. (2012) selected minimum morphological descriptors to distinguish passion fruit varieties, which indicated 22 of the 28 descriptors analyzed with a high contribution in the total observed variation. These results showed that the agronomic descriptors are useful to differentiate genetic resources of the genus *Passiflora*. This study aimed to characterize *Passiflora* spp. accessions and its genetic diversity based on quantitative morphoagronomic descriptors of flowers and fruits.

MATERIAL AND METHODS

The study was carried out at the Fruit Activity Support Unit and Laboratory of Food Analysis of Embrapa Cerrados, Planaltina-DF. The experimental design was a completely randomized design with 15 treatments (accessions) and four replicates, totaling 60 experimental plots. Each plot was composed of the average reading of 3 structures (descriptor of flower or fruit). Fifteen *Passiflora* spp accessions from the 'Flor da Paixão', Active Germplasm Bank (BAG) were characterized, namely: 1. *P. alata* (CPAC MJ-02-17), 2. *P. nitida* (CPAC MJ-01-03), 3. *P. suberosa* (CPAC MJ-35-02), 4. *P. caerulea* (CPAC MJ-14-01), 5. *P. hatschbachii* (CPAC MJ-50-01), 6. *P. maliformis* (CPAC MJ-58-01), 7. *P. quadrangularis* x *P. alata* (CPAC MJ-H-44), 8. *P. sidifolia* (CPAC MJ-16-02), 9. *P. malacophylla* (CPAC MJ-43-01), 10. *P. alata* (CPAC MJ-02-09), 11. *P. alata* (CPAC MJ-02-19), 12. *P. quadrangularis* (CPAC MJ-07-03), 13. *P. cincinnata* (CPAC MJ-26-03), 14. *P. alata* BRS Mel do Cerrado, 15. *P. tenuifila* BRS Vita.

The descriptors evaluated in flowers were androgynophore length (CAN), external diameter of the coronal cavity (DEEC), internal diameter of the coronal cavity (DIC), pedicel length (CPD), anther length (CA), anther width (LAN), ovary length (COV), ovary diameter (DOV). The descriptors evaluated in fruits were peel mass (MCA), seed mass (MSE), pulp mass (MPO), juice yield (RES), total titratable acidity (AT) and ratio between soluble solids and total titratable acidity (RATIO). Length, diameter and width data were obtained in centimeters (cm); mass data were obtained in grams (g).

The Mahalanobis distances (Mahalanobis, 1936) were estimated based on information from matrices of averages and covariates, calculated based on 14 morphoagronomic descriptors, eight of flowers and six of fruits, separately. Based on the matrices of distances, dendrogram cluster analysis was performed using the UPGMA method (*Unweighted Pair-Group Method using Arithmetic Averages*). Graphic dispersion was performed based on multidimensional scales using the main coordinates and the SAS software (SAS INSTITUTE INC., 2004) and Statistica software (STATSOFT INC., 2005). Pearson correlations were obtained based on the 14 morphoagronomic descriptors and on descriptors of each plant structure (flowers and fruits). The relative contribution of characters for divergence - SINGH (1981) was calculated with the aid of the Genes software (CRUZ, 2013). The stability of clusters was computed through the Bootstraping analysis with 500 replications through the Genes software

(CRUZ, 2006).

RESULTS AND DISCUSSION

Using the quantitative flower descriptors, the formation of seven groups was observed. Group one was similar to that formed using all descriptors, that is, with all *P. alata* accessions grouped together with CPAC MJ-H-44 hybrid (Figure 1). Corroborating the results observed, Ortiz et al. (2012) reported high genetic homogeneity when accessions of the same species are analyzed. Group two was composed of CPAC MJ-01-03, CPAC MJ-26-03, CPAC MJ-14-01 and CPAC MJ-58-01 accessions, which presented a very similar flower size and led to values numerically close and smaller genetic distances between them.

Group five was composed of CPAC MJ-16-02 and CPAC MJ-43-01 accessions. The other accessions, CPAC MJ-07-03, CPAC MJ-50-01 and *P. tenuifila* BRS Vita were not grouped together. Observing the dispersion graph, it was observed that CPAC MJ-07-03, CPAC MJ-50-01 and *P. tenuifila* BRS Vita accessions maintained the same tendency of distancing from the others, as well as CPAC MJ-35-02 accession, which belongs to subgenus *Decaloba*. Muschner et al. (2012) found genetic distance between subgenus *Decaloba* and subgenus *Passiflora*, confirming the classification in different subgenera. The other *Passiflora* spp. accessions were much closer to the dispersion chart, following the same dendrogram tendency.

For the quantitative fruit descriptors, the dendrogram shows the formation of three groups of similarity, and group one can be divided into two subgroups, the first composed of *P. alata* accessions and *P. alata* and *P. quadrangularis* hybrid and the second subgroup was composed of CPAC MJ-01-03, *P. tenuifila* BRS Vita, CPAC MJ-35-02, CPAC MJ-14-01 and CPAC MJ-43-01 accessions (Figure 2). Viana et al. (2010) verified a wide inter- and intraspecific morphological variation, obtaining a clear separation of species.

Group two was composed of CPAC MJ-50-01 and CPAC MJ-26-03 accessions. CPAC MJ-07-03 accession was not grouped with the other accessions. According to the dispersion chart, CPAC MJ-50-01 and CPAC MJ-26-03 accessions that appear together in the dendrogram were further separated, as was the CPAC MJ-07-03 accession. A large group of similarity was formed, as was observed in the accessions of group one of the dendrogram.

By the cluster analysis of 15 *Passiflora* spp. accessions using 14 quantitative morphoagronomic descriptors, the formation of 4 groups of similarity was verified (Figure 3). Considering all the evaluated characteristics, group one was composed of *P. alata* accessions and CPAC MJ-H-44 hybrid. Group two was the largest group, containing CPAC MJ-01-03, CPAC MJ-14-01, CPAC MJ-58-01, CPAC MJ-35-02, CPAC MJ-43-01, CPAC MJ-16 -02 and *P. tenuifila* BRS Vita accessions. Group three was composed of CPAC MJ-50-01 and CPAC MJ-26-03 accessions. CPAC MJ-07-03 accession did not form a group with other material probably due to its different size, considering the higher fruit weight and consequently a higher bark mass, higher pulp mass and higher juice yield. Observing the graphic dispersion, it was observed that accessions of group one and two are very close, forming a group with all accessions. Accessions that are together in the dendrogram forming group three in the dispersion are more distant from each other (Figure 3).

Correlations between estimates of genetic distances were obtained based on the descriptors of each plant structure (flowers and fruits) and on the 14 quantitative morphoagronomic descriptors. There was no significant correlation between genetic distances estimated based on fruit characteristics and those estimated based on flower characteristics (0.01^{ns}), evidencing that these characteristics are complementary for more complete inter- and intra-specific genetic diversity studies. There was a strong and highly significant correlation between genetic distances estimated based on all the descriptors and based on fruit descriptors (0.99^{**}), demonstrating that the fruit characteristics were determinant for the differentiation of accessions of species evaluated In this study. For the species evaluated in the present study, diversity estimates based solely on quantitative fruit characteristics would be sufficient to differentiate accessions.

Based on the method of Singh (1981), it was observed that ovary length (COV) was the variable that most contributed with the variability of accessions, with 25.14% (Table 1) for flower characteristics. Lawinscky et al. (2014) observed that the corona diameter was the variable that most contributed to the determination of the genetic diversity between *Passiflora alata* Curtis and *P. cincinnata* Mast species. Santos et al. (2011) found that the variables that most contributed to the genetic diversity among *P. foetida*, *P. sublanceolada* and the corresponding hybrid were flower diameter and peduncle length.

It was verified by the Singh method that variable peel mass (MCA) was the one that most contributed to the morphoagronomic characterization, with 51.30%. According to Santos et al. (2009), fruit

peel thickness is an important characteristic from the commercial point of view, since it is strongly related to juice yield, promoting an increase in the pulp percentage. After peel mass, the characteristics that most contributed to accession differentiation were Pulp mass (MPO) (28.21%) and total titratable acidity (AT) (17.98%). Pulp mass is directly related to the commercial value of passion fruits for the fresh market and for industrial processing that use pulp as main raw material. Total titratable acidity is a characteristic of great importance for the industry, since high acidity levels increase pulp conservation time and discourage the manifestation of microorganisms (NEGREIROS et al., 2008), being highly related to the quality of the beverage produced from the fruit pulp, which can be evaluated by the ratio between soluble solids and total titratable acidity (RATIO).

TABLE 1- Relative contribution of characters for divergence - SINGH (1981), androgynophore length (CAN), external diameter of the coronal cavity (DEEC), internal diameter of the coronal cavity (DIC), pedicel length (CPD), anther length (CA), anther width (LAN), ovary length (COV), ovary diameter (DOV), peel mass (MCA), seed mass (MSE), pulp mass (MPO), juice yield (RES), total titratable acidity (AT) and ratio between soluble solids and total titratable acidity (RATIO) of 15 *Passiflora* spp. accessions Embrapa Cerrados, Planaltina, DF, 2015.

Flower variables	Singh (%)	Fruit variables	Singh (%)
CAN (cm)	10.34	MCA (g)	51.30
DEEC (cm)	16.90	MSE (g)	0.26
DIC (cm)	2.40	MPO (g)	28.21
CPD (cm)	20.07	RES (%)	2.23
CA (cm)	5.28	AT	17.98
LAN (cm)	14.76	RATIO	0.02
COV (cm)	25.14		
DOV (cm)	5.01		

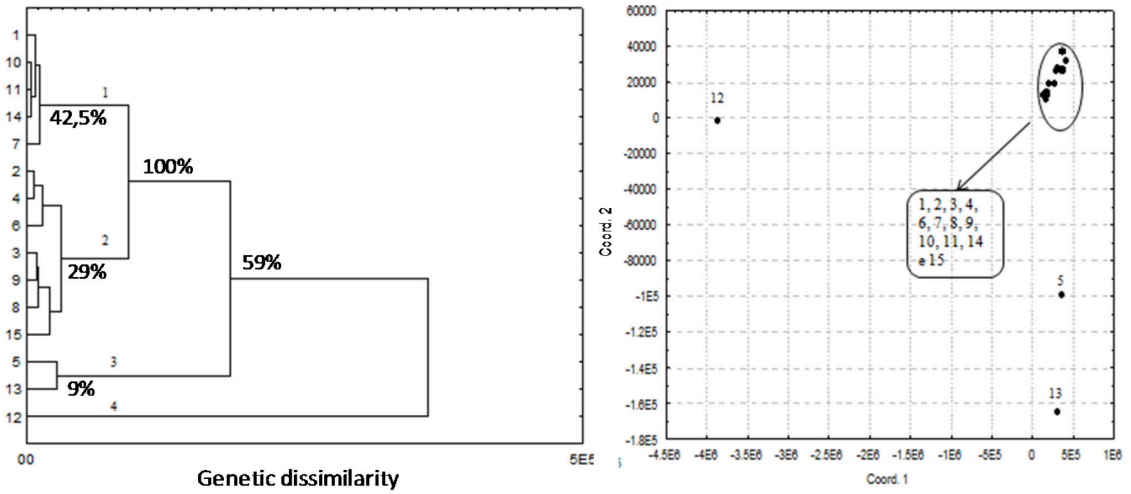

FIGURE 1- Clustering and dispersion analysis of 15 *Passiflora* spp. accessions based on the genetic dissimilarity matrix calculated using 8 quantitative flower morphological descriptors. The UPGMA method was used as a grouping criterion. The cophenetic correlation coefficient value (r) is 0.83. Embrapa Cerrados, Planaltina, DF, 2015.

Legend: 1. *P. alata* (CPAC MJ-02-17), 2. *P. nitida* (CPAC MJ-01-03), 3. *P. suberosa* (CPAC MJ-35-02), 4. *P. caerulea* (CPAC MJ-14-01), 5. *P. hatschbachii* (CPAC MJ-50-01), 6. *P. maliformis* (CPAC MJ-58-01), 7. *P. quadrangularis* x *P. alata* (CPAC MJ-H-44), 8. *P. sidifolia* (CPAC MJ-16-02), 9. *P. malacophylla* (CPAC MJ-43-01), 10. *P. alata* (CPAC MJ-02-09), 11. *P. alata* (CPAC MJ-02-19), 12. *P. quadrangularis* (CPAC MJ-07-03), 13. *P. cincinnata* (CPAC MJ-26-03), 14. *P. alata* BRS Mel do Cerrado, 15. *P. tenuifila* BRS Vita. Percentage numbers correspond to the stability of clusters computed by means of bootstrapping analysis with 500 replicates.

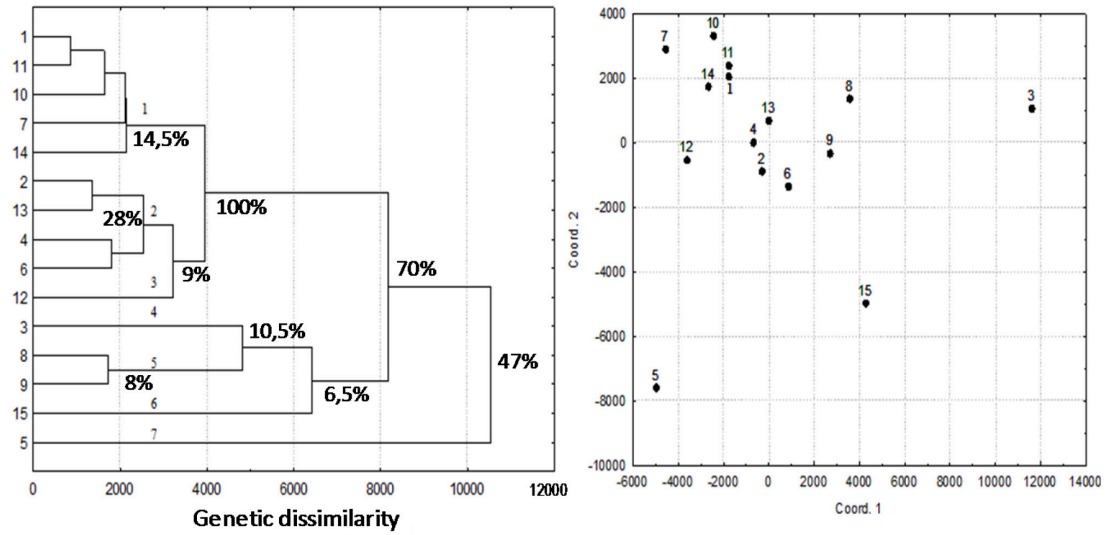

FIGURE 2- Clustering and dispersion analysis of 15 *Passiflora* spp. accessions based on the genetic dissimilarity matrix calculated using 6 quantitative flower morphological descriptors. The UPGMA method was used as a grouping criterion. The cophenetic correlation coefficient value (r) is 0.83. Embrapa Cerrados, Planaltina, DF, 2015.

Legend: 1. *P. alata* (CPAC MJ-02-17), 2. *P. nitida* (CPAC MJ-01-03), 3. *P. suberosa* (CPAC MJ-35-02), 4. *P. caerulea* (CPAC MJ-14-01), 5. *P. hatschbachii* (CPAC MJ-50-01), 6. *P. maliformis* (CPAC MJ-58-01), 7. *P. quadrangularis* x *P. alata* (CPAC MJ-H-44), 8. *P. sidifolia* (CPAC MJ-16-02), 9. *P. malacophylla* (CPAC MJ-43-01), 10. *P. alata* (CPAC MJ-02-09), 11. *P. alata* (CPAC MJ-02-19), 12. *P. quadrangularis* (CPAC MJ-07-03), 13. *P. cincinnata* (CPAC MJ-26-03), 14. *P. alata* BRS Mel do Cerrado, 15. *P. tenuifila* BRS Vita. Percentage numbers correspond to the stability of clusters computed by means of bootstrapping analysis with 500 replicates.

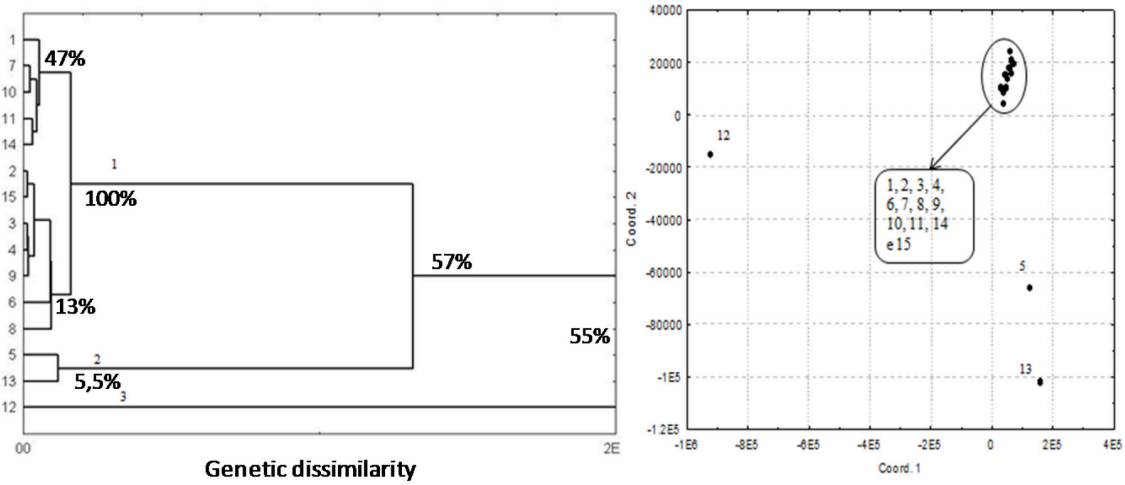

FIGURE 3- Clustering and dispersion analysis of 15 *Passiflora* spp. accessions based on the genetic dissimilarity matrix calculated using 14 quantitative flower morphological descriptors. The UPGMA method was used as a grouping criterion. The cophenetic correlation coefficient value (r) is 0.83. Embrapa Cerrados, Planaltina, DF, 2015.

Legend: 1. *P. alata* (CPAC MJ-02-17), 2. *P. nitida* (CPAC MJ-01-03), 3. *P. suberosa* (CPAC MJ-35-02), 4. *P. caerulea* (CPAC MJ-14-01), 5. *P. hatschbachii* (CPAC MJ-50-01), 6. *P. maliformis* (CPAC MJ-58-01), 7. *P. quadrangularis* x *P. alata* (CPAC MJ-H-44), 8. *P. sidifolia* (CPAC MJ-16-02), 9. *P. malacophylla* (CPAC MJ-43-01), 10. *P. alata* (CPAC MJ-02-09), 11. *P. alata* (CPAC MJ-02-19), 12. *P. quadrangularis* (CPAC MJ-07-03), 13. *P. cincinnata* (CPAC MJ-26-03), 14. *P. alata* BRS Mel do Cerrado, 15. *P. tenuifila* BRS Vita. Percentage numbers correspond to the stability of clusters computed by means of bootstrapping analysis with 500 replicates.

CONCLUSION

The morphoagronomic characterization based on quantitative descriptors of flowers and fruits contributed to the phenotypic differentiation among the 15 *Passiflora* spp. accessions, serving as an important instrument to quantify the variability among accessions. The fruit characteristics contributed more decisively to the differentiation of accessions evaluated in this study. The quantitative characterization of flower and fruit structures in a complementary way is important for more complete studies on the genetic characterization and genetic diversity of the genus *Passiflora*.

REFERENCES

CASTRO, J.A.; NEVES, C.G.; DE JESUS, O.N.; OLIVEIRA, E.J. Definition of morpho-agronomic descriptors for the characterization of yellow passion fruit. **Scientia Horticulturae**, The Hague, v.145, n.1, p.22-17, 2012.

CRUZ, C.D. Genes - a software package for analysis in experimental statistics and quantitative genetics. **Acta Scientiarum Agronomy**, Maringá, v.35, n.3, p.271-276, 2013.

FALEIRO, F. G.; JUNQUEIRA, N. T. V.; BRAGA, M. F. **Maracujá: demandas para a pesquisa.** Planaltina: Embrapa Cerrados, 2006, 54 p.

FALEIRO, F.G.; JUNQUEIRA, N.T.V.; BRAGA, M.F.; COSTA, A.M. **Conservação e caracterização de espécies silvestres de maracujazeiro (*Passiflora* spp.) e utilização potencial no melhoramento genético, como porta-enxertos, alimentos funcionais, plantas ornamentais e medicinais - resultados de pesquisa.** Planaltina: Embrapa Cerrados, 2012. 34 p. (Documentos, 312)

FALEIRO, F.G.; JUNQUEIRA, N.T.V.; BRAGA, M.F.; PEIXOTO, J.R.**Caracterização de germoplasma e melhoramento genético do maracujazeiro assistidos por marcadores moleculares: resultados de pesquisa 2005-2008**. Planaltina: Embrapa Cerrados, 2008. 59 p. (Boletim de Pesquisa e Desenvolvimento, 207)

LAWINSCKY, P.R.; SOUZA, M.M.; BELO, G.O.; VIANA, A.J.C.; MELO, C.A.F.; OLIVEIRA, C.S.L. Morphological characterization and genetic diversity in *Passiflora alata* Curtis and *P. cincinnata* Mast. (Passifloraceae). **Brazilian Journal Botany**, São Paulo, v.37, n.3, p.272-261, 2014.

MAHALANOBIS, P.C. On the generalized distance in statistics. **Proceedings of The National Institute of Sciences of India**, New Dehli, v.12, p.49-55, 1936.

MUSCHNER, V.C.; ZAMBERLAN, P.M.; BONATTO, S.L.; FREITAS, L.B. Phylogeny, biogeography and divergence times in *Passiflora* (*Passifloraceae*). **Genetics and Molecular Biology**, Ribeirão Preto, v.35, n.4, p.1043-1036 2012.

NEGREIROS, J.R.S.; ARAÚJO, S.E.; ÁLVARES, V.S.; LIMA, V.A.; OLIVEIRA, T.K. Caracterização de frutos de progênies de meios-irmãos de maracujazeiro-amarelo em Rio Branco – Acre. **Revista Brasileira de Fruticultura**, Jaboticabal, v.30, n.2, p.437-431, 2008.

ORTIZ, D.; BOHÓRQUEZ, A.; DUQUE, M.C.; TOHME, J.; CUELLAR, D.; MOSQUERA, T. Evaluating purple passion fruit (*Passiflora edulis* Sims f. edulis) genetic variability in individuals from commercial plantations in Colombia. **Genetic Resources and Crop Evolution**, Dordrecht, v.59, n.6, p.1099-1089, 2012.

SANTOS, C.E.M.; BRUCKNER, C.H.; CRUZ, C.D.; SIQUEIRA, D.L.; PIMENTEL, L.D. Características físicas do maracujá-azedo em função do genótipo e massa do fruto. **Revista Brasileira de Fruticultura**, Jaboticabal, v.31, n.4, p.1110-1102, 2009.

SANTOS, E.A.; SOUZA, M.M.; VIANA, A P.; ALMEIDA, A.A.F.; FREITAS, J.C.O.; LAWINSCKY, P.R. Multivariate analysis of morphological characteristics of two species of passion flower with ornamental potential and of hybrids between them. **Genetics and Molecular Research**, São Paulo, v.10, n.4, p.2471-2457, 2011.

SAS INSTITUTE. **SAS user's guide**: statistic: version 9.1.3. Cary: SAS Institute, 2004. 846 p.

SINGH, D. The relative importance of characters affecting genetic diversity. **Indian Journal of Genetics and Plant Breeding**, New Delhi, v. 41, n. 2, p.245-237, 1981.

STATSOFT. **Statistica for windows (data analysis software system). Version 7.1**. Tulsa: Statsoft, 2005.

VIANA, A.J.C.; SOUZA, M.M.; ARAÚJO, I.S.; CORRÊA, R.X. Genetic diversity in *Passiflora* species determined by morphological and molecular characteristics. **Biologia Plantarum**, Praha, v.54, n.3, p.538-535, 2010.