MORPHOMETRIC DESCRIPTORS AND PHYSIOLOGICAL SEED QUALITY FOR SELECTING Aspidosperma pyrifolium Mart. MATRIX TREES¹

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ABSTRACT - Aspidosperma pyrifolium Mart. (Apocynaceae) is a tree species with high ecological and economic potential. Therefore, the need to select trees for producing high-quality seeds is evident. In this sense, the objective of this study was to select *A. pyrifolium* matrices based on the morphometric descriptors of fruits and seeds and the physiological quality of seeds from a natural population. For this, 11 *A. pyrifolium* trees were selected, and their fruits and seeds were submitted to biometric analysis; in addition, the physiological quality of the seeds was evaluated. The morphometric characteristics of fruits and seeds and the physiological quality of the seeds evidenced differences and variations among the seeds of the different *A. pyrifolium* trees, making it possible to group them according to the similarity degree. Thus, seven trees were selected as seed matrices based on superior physiological quality and genetic dissimilarity.

Keywords: Biometry. Genetic diversity. Dry forests. Pereiro tree.

DESCRITORES MORFOMÉTRICOS E QUALIDADE FISIOLÓGICA DE SEMENTES PARA SELEÇÃO DE MATRIZES DE *Aspidosperma pyrifolium* Mart.

RESUMO - *Aspidosperma pyrifolium* Mart. (Apocynaceae) é uma espécie arbórea com elevado potencial ecológico e econômico, por isso, evidencia-se a necessidade de seleção de árvores para a produção de sementes de alta qualidade. Nesse sentido, o objetivo deste estudo foi selecionar matrizes de *A. pyrifolium* com base nos descritores morfométricos de frutos e de sementes e na qualidade fisiológica de sementes provenientes de uma população natural. Para isso, onze árvores de *A. pyrifolium* foram selecionadas, cujos frutos e sementes foram submetidos à análise biométrica, bem como realizada a avaliação da qualidade fisiológica das sementes. Os caracteres morfométricos de frutos e de sementes, e a qualidade fisiológica das sementes evidenciaram diferenças e variações entre as sementes oriundas das diferentes árvores de *A. pyrifolium*, tornando possível agrupá-las conforme o grau de similaridade. Assim, foram selecionadas sete árvores como matrizes produtoras de sementes baseado na qualidade fisiológica superior e na maior dissimilaridade genética.

Palavras-chave: Biometria. Diversidade genética. Florestas secas. Pereiro.

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¹Received for publication in 10/31/2018; accepted in 03/22/2019.

Paper extracted from the undergraduate thesis of the first author.

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INTRODUCTION

Aspidosperma pyrifolium Mart. (Apocynaceae) is a tree species of economic and ecological importance and with ample uses; under favourable environmental conditions, it can reach a height of 7 to 8 meters, with a high capacity for regenerating degraded areas (MAIA, 2004). The species is also important because of the different uses of its wood, either for construction, woodworking or for the production of firewood (CHAVES et al., 2014); in addition, it is widely used in traditional and veterinary medicine (COSTA JUNIOR et al., 2014; SILVA et al., 2014).

The fruits are woody and dehiscent, containing winged seeds that are dispersed by the wind (SILVA et al., 2014); the fruit and seed extracts have antioxidant potential in inflammatory (LIMA et al., 2017) and neurodegenerative diseases (ARAÚJO et al., 2018). In addition, the plant species of the northeastern semiarid region are important to support the economic viability of the agricultural sector (ARAÚJO et al., 2014).

The over-exploitation of native tree species has been considered as one of the factors that contribute most to the increase in degraded areas (SANTOS et al., 2014), which results in the decline of forest biodiversity (DANTAS et al., 2008). For this reason, selecting matrix trees has a relevance for producing seeds destined to establish quality seedlings, aiming at programs for forest restoration, reforestation, recovery of degraded areas, urban afforestation and species preservation.

Environmental problems have intensified the interest in the propagation of native species (SILVA; CARVALHO, 2008), and the seeds used for this purpose should represent the genetic variability of the population, thereby avoiding inbreeding and preserving the long-term evolutionary potential of the species (SEBBENN, 2002). In addition, the physiological quality of the seeds is an important factor to ensure germination performance under field conditions and to conserve their physiological potential during storage.

Physiological quality is evaluated through viability and vigour tests; among the tests associated with germination, the first count, germination speed index, growth analysis and seedling dry mass allocation, as well as seedling emergence under unfavourable germination conditions may be highlighted (MARCOS FILHO, 2015).

Biometric characterisation of fruits and seeds enables the estimation of intra- and interpopulation genetic variability (VIEIRA; GUSMÃO, 2008). Therefore, studies on the morphometry of native forest species, aiming at the variability among matrices, are crucial. In this sense, analysis of the genetic population diversity and the evaluation of the physiological quality of the seeds from different trees in natural populations are relevant to select matrices and areas for seeds that meet the quality requirements demanded by the conservation programs.

Thus, the objective of this study was to select *A. pyrifolium* matrices based on the morphometric descriptors of fruits and seeds and on the physiological quality of seeds from individuals of a natural population of *A. pyrifolium*.

MATERIAL AND METHODS

Study location and material

The study area was located at the *Fazenda Manoel de Souza* (7° 09' 40.8" S and 36° 18' 25.8" W), in the rural area of Soledade/PB, Brazil, in the Curimataú microregion, Agreste region. The climate is predominantly hot and dry; according to the Köppen classification, it is semi-arid subtype BS'h, with a mean annual precipitation of 500 mm, a temperature between 16.7 and 31.0°C and a relative humidity of 65%.

In the study population (16.5 ha), 11 *A. pyrifolium* trees with good phytosanitary appearance, well-shaped crowns and high fruit yields were selected. The selected trees were marked, georeferenced and had their fruits manually collected using pruning shears. The material collected from each tree was submitted to fruit and seed evaluations.

Morphometric descriptors of fruit and seeds; seed quality

Length (mm), width (mm) and thickness (mm) measurements of 70 fruits and 100 seeds without wings of each tree were obtained with a digital calliper (0.01 mm), and fresh fruit and seed masses (mg) were obtained by weighing on a precision analytical scale (0.001 g). The number of seeds per fruit was also counted.

The following evaluations were performed to determine the physiological quality of the seeds of each tree:

Moisture content (MC): Fifteen seeds (2.0 mg) of each tree were oven-dried at 105 ± 3 °C for 24 h, and the results were expressed as percentage (BRASIL, 2009).

Germination (G): The experimental design was completely randomised with four replicates of 50 seeds per tree, which were immersed in detergent solution (5 drops/100 mL of water) for 5 min, withdrawn and immersed again in 2% sodium hypochlorite solution for 2 min; subsequently, they were rinsed with distilled water (BRASIL, 2013). Afterwards, the seeds were placed between three sheets of paper towel (Germitest[®]) moistened with distilled water in the proportion of 2.5 times its dry weight. Finally, the leaves were organised in roller systems, conditioned in transparent plastic bags and

incubated in a Biochemical Oxygen Demand (BOD) germinator regulated at 25°C, with a 12-h photoperiod. The number of normal seedlings (BRASIL, 2013) was then determined on the 13th day after sowing, and the results were expressed as percentage.

The first count (PC) was carried out in conjunction with the germination test, counting the number of seeds germinated on the 3^{rd} day after sowing; the results were expressed as percentage.

Length (L) and dry mass (DM) of seedlings: normal seedlings (BRASIL, 2013) resulting from the germination test were measured (apical bud until primary root apex) with a ruler graduated in millimetres, and the results were expressed in cm.seedling⁻¹. Subsequently, the cotyledons were removed, and the seedlings were placed in paper bags, air-dried at 80°C for 24 h and weighed on an analytical scale (0.001 g); the results were expressed in mg.seedling⁻¹ (KRZYZANOWSKI; VIEIRA; FRANÇA-NETO, 1999).

Seedling emergence (E): the experimental design was a randomised block consisting of four blocks with 50 seeds for each tree, with the test being conducted in experimental beds in full sun, using sand as substrate. The seeds were seeded with the embryonic axis region facing down to a depth of 2.0 cm, and the number of emerging seedlings (emergence of the cotyledon above the substrate) was determined until the counts were constant (26 days); the results were expressed as percentage.

Germination speed index (GSI) and seedling emergence speed index (ESI): data on the daily counting of the number of germinated seeds and emerged seedlings, respectively, were applied in the formula proposed by Maguire (1962).

Data analysis

The data for the fruit and seed morphometric descriptors and the results of the tests to evaluate the physiological quality of the seeds were submitted to the Lillefors normality test (at the 1% probability level). As the data met the assumptions for parametric statistical analysis, they were submitted to ANOVA, and the means of the treatments were compared using the Scott-Knott test at 5 and 1% probability; the analyses were performed using the

software package Assistat[®], version 7.7 beta.

The genetic, phenotypic and environmental variance parameters related to the contribution of fruit and seed morphometric characteristics, as well as heritability (h^2) and the relative contribution of these traits based on Singh (1981) divergence, were evaluated using the Genes[®] program.

The genetic distance between the individuals was evaluated using BioEstat[®] version 5.3, and multivariate analysis (Euclidian distance with standardised mean) was applied based on the morphometric descriptors. A dendrogram was created by Cluster analysis from the distance matrix by the Unweighted pair-group method, using the arithmetic average (UPGMA) method.

RESULTS AND DISCUSSION

Physiological seed quality

The moisture content of the seeds from the 11 A. pyrifolium trees was 1.0%, except for A88 (GU = 8.7%), in which the variation was 2.3% when compared to the lowest value (A97 = 6.4%) (Table 1). This variation among seeds is within the maximum allowed limit of 2.0%, which is necessary to patronise the evaluations and to obtain consistent results in the vigour tests (MARCOS FILHO, 2015). Small differences have also been observed in seeds from different *Erythrina velutina* Willd trees (GUEDES et al., 2009) and *Poincianella pyramidalis* (Tul.) L. P. Queiroz (LIMA et al., 2014). Seeds with high moisture content can degrade their reserves more quickly due to an increased respiratory activity, resulting in reduced vigour.

Seeds from A89, A90, A95 and A97 trees showed higher germination rates, while those from A93 and A96 trees presented intermediate performance, and the others produced lower-quality seeds (Table 1). Variations in the physiological quality of seeds from different trees have also been observed for *Tabebuia chrysotricha* (Mart. Ex A. DC.) Standl. (SANTOS et al., 2009), *P. pyramidalis* (LIMA et al., 2014) and *Ceiba speciosa* St. Hil (ROVERI NETO; PAULA, 2017), evidencing that individuals of the same population produce seeds with different viabilities.

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Tree	MC (%)	G ** (%)	FGC ** (%)	GSI **	L ^{**} (cm.seedling ⁻¹)	DM ** (mg.seedling ⁻¹)	E ** (%)	ESI *
A88	8.7	45 d	19 d	7.78 c	7.25 c	265.6 d	25 b	1.13 b
A89	7.2	96 a	69 a	14.19 a	9.68 a	681.8 a	60 a	2.40 a
A90	6.9	93 a	56 b	14.90 a	8.20 b	725.2 a	57 a	2.26 a
A91	7.4	69 c	46 b	11.57 b	7.20 c	356.0 c	45 a	1.99 a
A92	7.0	72 c	54 b	11.44 b	6.51 d	363.3 c	32 b	1.32 b
A93	6.9	83 b	35 c	10.36 b	7.97 b	501.4 b	41 a	1.68 a
A94	6.7	48 d	31 c	7.10 c	6.05 d	221.3 d	14 b	0.51 b
A95	6.6	97 a	49 b	13.34 a	9.01 a	714.7 a	51 a	1.99 a
A96	6.8	86 b	73 a	13.72 a	10.17 a	697.9 a	48 a	1.95 a
A97	6.4	95 a	24 d	10.86 b	9.29 a	565.8 b	51 a	2.12 a
A98	6.9	67 c	48 b	9.85 b	6.04 d	346.5 c	30 b	1.35 b
CV (%)	-	9.82	17.94	11.75	9.43	13.23	36.21	42.18

Table 1. Moisture content (MC), germination (G), first germination count (FGC), germination speed index (GSI), length (L), dry mass (DM), emergence (E) and emergence speed index (ESI) of seedlings of 11 *A. pyrifolium* trees.

Means followed by the same letter in the column did not differ statistically from one another by the Scott -Knott test at the 5% probability level (p < 0.05) *and at 1% probability (p < 0.01)**.

The trees A89 and A96 produced seeds with the highest germination percentages (69 and 73%, respectively) on the third day after sowing, followed by those from A90, A91, A92, A95 and A98 trees, which demonstrated intermediate performance; superior GSI results were also observed for seeds from A89, A90, A95 and A96 trees, followed by A91, A92, A97 and A98 with intermediate values (Table 1).

Seeds belonging to A89, A95, A96 and A97 trees showed a higher seedling growth capacity, followed by A90 and A93 trees, and the dry matter quantification highlighted A89, A90, A95 and A96 trees (Table 1). Thus, seedling length and dry mass tests were able to rank seeds from *A. pyrifolium* trees at different vigour levels. In a study on the physiological quality of seeds from different *P. pyramidalis* matrices, the dry mass quantification of seedlings was efficient in discriminating seed vigour from different lots (LIMA et al., 2014).

The percentage of emerged seedlings and the ESI were higher for the seeds from A89, A90, A91, A93, A95, A96 and A97 trees than for those from the other trees (Table 1). The emergence test is considered efficient to differentiate seed lots at

different levels of physiological quality by subjecting them to adverse field conditions, where it is assumed that only more vigorous seeds will emerge (MARCOS FILHO, 2015) and be further likely to establish a rapid and uniform stand.

Therefore, the vigour tests used to complement the germination test results were able to classify the seeds from the different *A. pyrifolium* trees into different vigour levels, highlighting the A89, A90, A95 and A96 trees as producers of quality seeds due to their better performance in most of the observed variables and the A91, A93 and A97 trees as intermediate-quality seed producers, which presented superior performance in the emergence and ESI tests in relation to the other seed lots.

Morphometric descriptors of fruits and seeds

There was a significant difference between the physical characteristics of the fruits (Table 2) and the seeds (Table 3) from the different *A. pyrifolium* trees, with their variations in dimensions and fresh mass of the fruits and seeds possibly being related to phenotypic influences.

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Tree Mass (g)		Length (mm)	Width (mm)	Thickness (mm)	No. of seeds (per fruit)	
A88	15.459 d	63.80 e	41.03 e	11.25 d	8.10 b	
A89	13.712 e	62.32 f	37.77 f	11.14 d	7.12 d	
A90	19.652 c	73.54 c	44.74 c	13.73 b	7.62 c	
A91	11.454 f	61.55 f	40.27 e	12.66 c	7.98 b	
A92	19.479 c	73.27 с	42.22 d	14.68 a	8.03 b	
A93	15.553 d	68.20 d	43.19 d	13.267 b	7.36 d	
A94	24.070 a	85.88 a	47.05 b	13.19 b	7.64 c	
A95	18.739 c	78.34 b	43.32 d	12.31 c	7.15 d	
A96	19.881 c	68.34 d	46.08 b	14.84 a	7.73 c	
A97	21.444 b	75.76 b	44.37 c	13.64 b	7.68 c	
A98	20.774 b	65.19 e	50.02 a	14.39 a	8.69 a	
CV (%)	4.20	2.64	1.97	5.66	3.65	

Table 2. Mean values of length, width, thickness and mass of fruits, as well as number of seeds per fruit of 11 *A. pyrifolium* trees.

Averages followed by the same letter did not differ statistically from each other by the Scott-Knott test at the 5% probability level (p < 0.05).

The number of seeds formed by the *A. pyrifolium* fruit presented a smaller variation compared to the other analysed variables. Tree A98 produced larger fruits (50.02 mm) and had the highest number of seeds per fruit (Table 2). Although it is characteristic of each species, the number of seeds formed by the fruit can be affected by environmental factors depending on the tree

location (SANTOS et al., 2009), while the morphometric characteristics of the seeds can vary within the same population due to genetic diversity (FREIRE et at., 2015). The relief of the seed collection region and the soil and climate conditions were homogeneous; therefore, the greater variation can be attributed to the genetic variability among the trees.

Table 3. Mean values of length, width, thickness and fresh mass of the seeds from 11 A. pyrifolium trees.

Tree	Length (mm)	Width (mm)	Thickness (mm)	Mass (mg)
A88	17.25 a	15.0 a	0.77 a	88.5 b
A89	15.14 c	13.34 b	0.75 a	97.6 a
A90	17.17 a	15.38 a	0.61 b	109.1 a
A91	14.86 c	12.31 b	0.67 a	66.5 b
A92	15.11 c	13.01 b	0.76 a	76.9 b
A93	14.29 d	13.79 b	0.51 c	96.1 a
A94	15.84 b	16.29 a	0.44 c	78.7 b
A95	17.13 a	15.19 a	0.68 a	107.9 a
A96	17.42 a	16.26 a	0.58 b	109.8 a
A97	16.84 a	16.40 a	0.55 b	89.2 b
A98	16.07 b	14.86 a	0.43 c	87.3 b
CV (%)	2.13	8.23	10.65	13.02

Means followed by the same letter did not differ statistically from each other by the Scott-Knott test at the 5% probability level (p < 0.05).

The A95 tree presented seeds with higher dimensions and fresh mass compared to seeds from the other trees, while the A88 tree presented seeds with similar values for length, width and thickness; seeds from A90 and A96 trees showed similar values for length, width and fresh mass. The seeds of A97 demonstrated superiority only for length and width (Table 3).

Although the trees were within the same natural population, these variations in size may result from population genetic variability or from environmental factors such as changes in temperature, resource availability, rainfall index, day length, phenotype of each individual and other factors which influence the expression of certain characteristics of the species (GUSMÃO; VIEIRA; FONSECA, 2006; SANTOS et al., 2009).

The variation observed for the morphometric characteristics of fruits and seeds from *A. pyrifolium* trees is due to genotypic variation (except for fruit thickness). Thus, these characteristics have high heritability ($h^2 > 95\%$), except for number of seeds per fruit (77.2%) and seed width (48.6%) (Table 4). The percentage heritability values express the phenotypic variance in proportional terms, resulting from the genotypic variation related to the evaluated metric components (fruit and seed biometry) (SILVA et al., 2007).

Table 4. Genotypic, phenotypic, environmental and heritability parameters related to the contribution of morphometric characteristics of fruits and seeds of *A. pyrifolium*.

Characteristic		h^2	CGV		
Characteristic	Phenotypic	Environment	Genotypic	(%)	(%)
Fruit					
Mass **	15.384	0.219	15.164	98.6	21.4
Length	62.464	1.231	61.233	98.0	11.1
Width **	13.158	0.307	12.851	97.7	8.2
Thickness ^{ns}	58.674	60.394	0.000	-	-
No. of seeds per fruit **	0.175	0.040	0.135	77.2	4.8
Seeds					
Length **	1.128	0.030	1.099	97.4	6.4
Width *	2.821	1.449	1.372	48.6	7.7
Thickness **	0.022	0.001	0.022	97.7	23.9
Mass **	0.241	0.004	0.237	98.2	17.3

^{ns}not significant; ^{*}, ^{**}significant at the 5 and 1% probability levels, respectively. h^2 : heritability; CGV: coefficient of genetic variation.

In a previous study, differences have also been observed for *C. speciosa* fruits and seeds among he matrix trees for all evaluated biometric characteristics, where the phenotypic value is associated with the genotype of the trees, since the biometric characteristics are strictly genetically controlled and have low environmental influence (ROVERI NETO; PAULA, 2017). characteristics that contribute most (50.1%) to the genotypic variation results according to the Singh (1981) divergence, whereas the number of seeds per fruit, seed width and fruit thickness contributed only slightly (1.9%) to explain the total variation (Table 5). The *C. speciosa* seed mass also presented a greater variation in relation to the other biometric aspects (ROVERI NETO; PAULA, 2017).

Fruit and seed mass are the morphometric

Table 5. Relative contribution of the morphometric characteristics of *A. pyrifolium* fruits and seeds to the divergence of Singh (1981).

Characteristic	Percentage (%)
Fruit mass	30.00
Seed mass	20.08
Seed thickness	14.25
Seed length	13.03
Fruit length	11.80
Fruit width	8.98
No. of seeds in per fruit	1.02
Seed width	0.57
Fruit thickness	0.29
Total	100.0

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Intrapopulation genetic divergence

The data on the physical characterisation of fruits and seeds, as well as the number of seeds per fruit, were used to elaborate the Euclidean distance

matrix among the *A. pyrifolium* individuals. Thus, the smallest and largest distances were between A90 and A96 (1.1488) and between A91 and A94 (6.7939), respectively (Table 6).

Table 6. Euclidean distance matrix among 11 A. pyrifolium trees based on the morphometric descriptors of fruits and seeds.

	A88	A89	A90	A91	A92	A93	A94	A95	A96	A97	A98
A88											
A89	2.9996										
A90	4.0017	4.5359									
A91	3.8130	2.6752	5.9611								
A92	4.5156	4.4076	4.0549	4.5668							
A93	4.4423	3.2355	3.4720	4.0673	3.7269						
A94	5.5288	6.1011	3.7368	6.7939	4.5387	4.2994					
A95	5.0044	4.8074	3.0582	6.7151	5.3561	4.6349	4.3515				
A96	4.4722	5.3083	1.1488	6.6129	4.6051	4.0417	3.9781	3.9236			
A97	3.9608	4.8826	1.8900	5.9025	3.9271	3.6492	2.2401	3.4993	2.0818		
A98	5.0514	5.8637	3.4519	6.2021	4.0765	3.7159	3.5576	5.8896	3.1880	3.0322	

The Euclidean distance matrix based on the morphometric descriptors of *A. pyrifolium* fruits and seeds was used for elaborating the similarity dendrogram (UPGMA). Cluster analysis at the 60% level demonstrates the formation of two groups: the

first composed of A89 and A91 trees and the second of A90, A96 and A97 trees, while the other individuals were more dissimilar and did not form clusters (Figure 1).

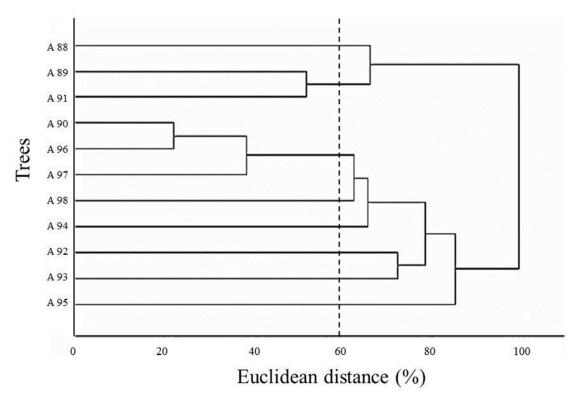


Figure 1. Grouping analysis of 11 *A. pyrifolium* individuals based on the morphometric descriptors of the fruits and seeds for the Euclidian distance.

Analysis of the nine morphometric descriptors of the 11 studied trees enabled differentiating them among themselves, as well as

grouping the closest ones regarding the similarity between them. Thus, based on this analysis, combined with the physiological test results, it is

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possible to identify and select A89, A90, A91, A93, A95, A96 and A97 trees as seed-producing matrices with high physiological quality and satisfactory genetic variability.

The variation found among the trees, based on the morphometric descriptors of the fruits and seeds, evidences that the matrices do not present a high degree of similarity, even though they belong to the same sampling point, indicating genetic variability. Species that are isolated from the native vegetation are not able to perform gene exchange between individuals, which results in species loss and important characteristics of medium-and longterm survival (GUOLLO; FELIPPI; POSSENTI, 2016).

A similar result has been observed in a genetic variability study among *Mimosa scabrella* Benth. based on seed analysis, which found a high variation between biometric characteristics of seeds from different trees and provenances (MENEGATTI et al., 2017). Intrapopulational genetic divergence has also been observed through morphometric descriptors in *Casearia grandiflora* Camb. populations, with most of the variations being observed within the populations (COSTA et al., 2016).

Genotypic variations among individuals of a species within a single population provide essential subsidies for improving a characteristic of interest, constituting an important parameter for biotechnology, with an emphasis on plant breeding as well as establishing active germplasm banks (SANTOS et al., 2009; SILVA et al., 2010). Thus, research with this focus enables selecting characteristics of interest that meet the needs of commercial nurseries and recovery programs for degraded areas with seeds that promote the establishment of vigorous plants and meet the requirements of desired genetic variability.

Considering the above, variations in the studied physical characteristics of *A. pyrifolium* matrix fruits and seeds reveal the potential of the population for selecting seed-producing matrices. The genetic variability among the matrices also suggests the potential of the population for collecting seeds to produce seedlings destined to programs for the recovery of degraded areas, reforestation, urban tree planting or even to guarantee the conservation of this species. Thus, the maximum representativeness of the genetic variability of the species is necessary to meet these objectives and to avoid inbreeding.

CONCLUSIONS

The morphometric characteristics of fruits and seeds and the physiological quality of seeds show differences and variations among the seeds from different *A. pyrifolium* trees, making it possible to group them according to the degree of similarity. Thus, 7 of the 11 initial trees can be selected as seedproducing matrices based on superior physiological quality and genetic dissimilarity.

ACKNOWLEDGEMENTS

This study was financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior - Brasil (CAPES) - Finance Code 001.

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