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Physical and fruit region characterization of araticum and seed germination as a result of accesses

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Abstract

The physical characteristics of the fruits are of great importance for the identification and selection of superior genetic materials, appropriate for commercialization or industrial use. Given the above, the objective was to physically characterize the araticum fruit and to verify the influence of accessions and fruit region under seed germination. The work was developed at Unioeste, Marechal Cândido Rondon Campus (PR), and conducted at the University Laboratory of Post-Harvest Technology. Ripe fruits of araticum (*Annona sylvatica*) were collected from four native plants (accessions A1, A 2, A3, and A4) at the Experimental Farm of the University in February 2019. As for the physical evaluation of the fruits, four repetitions of 10 randomly chosen ripe fruits per access were evaluated. The longitudinal diameter, transversal diameter, fruit biomass, epicarp biomass, fruit pulp, number of seeds, and mass of 100 seeds were the characteristics evaluated. The experimental design used for the germination test was completely randomized, in a factorial scheme 4 x 2 [4 accessions (A1, A2, A3, and A4) x 2 regions of the fruit (proximal and distal) in 500 mg L^{-1} of acid gibberellic (GA3)], containing 4 repetitions and 25 seeds per repetition. The evaluations were carried out from the 15th day of the experiment setup until 105 days. The characteristics evaluated were: first germination count (%), germination percentage (%), germination speed index, and average germination time (days). The fruits had an average biomass of 63.41 g, an average of 38 seeds per fruit, and fruit pulp with an average of 39.63 g. The germination of araticum seeds was not influenced by the accessions and fruit region.

Keywords: *Annona sylvatica* (A. St.-Hil) Mart., native species, postharvest, seed vigor

Introduction

The Annonaceae family has about 120 genera and more than 2,300 species. Five genera are of economic importance (Braga Sobrinho, 2014), and one of the most representative is *Annona* (Nunes et al., 2012). Araticum [*Annona sylvatica* (A. St. – Hil.) Mart.] is a native species of the Atlantic Forest biome, which, in Brazil, is predominant from the state of Pernambuco to Rio Grande do Sul (Lorenzi, 2016) and has economic potential in the fruit-growing sector. Besides its economic value, this species is crucial for forest restoration in degraded areas due to its high environmental adaptability (Lorenzi, 2016).

The Brazilian agroindustrial sector has growing potential for exploitation of native fruits (Hansen et al., 2013). Despite its great potential, only locals consume araticum fresh or in juices, ice cream, cakes, sweets, jellies, among others (Arruda et al., 2016). Therefore, studies detailing the characteristics of this fruit and its uses in the food industry are needed, so that national production could be maximized.

Fruit physicochemical characteristics are crucial to identify and select superior genetic materials to produce higher-quality fruits, which are suitable for in natura marketing or industrial use (Carvalho et al., 2013; Oliveira et al., 2010).

Fruits must be physically and chemically characterized so that higher-quality accessions are identified and adapted to the growing region (Greco et al., 2014). Despite the relevant number of studies on some native species (Wu et al., 2013), most of the information on physical-chemical and nutritional compositions are still scarce.

In addition to fruit, seed, and seedling physical characterization, sexual reproduction and seed position in the fruit shall also be investigated. Such information is still lacking for forest species (Mendonça et al., 2016).

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Although a few studies have already verified the effect of seed position in the fruit on the germination of different species such as *Mimosa caesalpiniifolia* Benth. (Freitas et al., 2013), *Enterolobium contortisiliquum* (Vell.) Morong (Lessa et al., 2014), and *Annona macroprophyllata* Donn. Sm (González-Esquinca et al., 2015) no research has assessed it for *A. sylvatica*.

Given the above, the present study aimed to characterize araticum fruits physically and verify the effect of accessions and seed position in the fruit on seed germination.

Material and Methods

The experiment was carried out at the Laboratory of Post-Harvest and Seed Technology, State University of Western Paraná (UNIOESTE), Campus Marechal Cândido Rondon (Paraná State), Brazil. To do so, ripe fruits of araticum (*Annona sylvatica*) were picked from four native plants (accessions: A1, A2, A3, and A4) in February 2019. These plants were in the Experimental Farm of the Nucleus of Experimental Stations of the UNIOESTE university.

According to Köppen, the local climate is classified as *Cfa*, which stands for subtropical with mean temperatures below 18 °C (mesothermal) and above 22 °C during the coldest and warmest months, respectively (Alvares et al., 2013). There, summers are warm and winters present infrequent frosts. Rains tend to concentrate in the summer months but without a well-defined dry season. Average yearly rainfall ranges from 1,600 to 1,800 mm (Caviglione et al., 2000).

With the aid of a pruning knife, the fruits were collected from native trees, which are about 15-20 m high and phytosanitary healthy. These trees were in a natural site belonging to the UNIOESTE. The plants were spaced about 5 to 20 meters apart, and the fruits were collected based on their color (yellow). The study comprised two trials.

The first experiment evaluated fruit and seed physical characteristics and had 4 replications of 10 ripe fruits per accession (A1, A2, A3, and A4), which were randomly selected. The variables longitudinal diameter (LD) and transverse diameter (TD) were measured with the aid of a digital caliper, and results expressed in mm per fruit. Fruit biomass (FB) and epicarp biomass (EB) were measured using a digital scale with a capacity for six kilograms and accuracy of 0.002 g, and results expressed in g per fruit. After the fruits were pulped, the pulp (FP) was weighed on a digital scale with an accuracy of 0.001g, and results expressed in g per fruit. Lastly, the number of seeds per fruit (NS) was counted manually and 1,000-seed mass (TSM; in g) determined according to the Rules for Seed Analysis. After collection, the fruit epicarp (i.e., fruit peel) was removed, placed into 5-L plastic buckets, and then left to rest for 48 h. Afterwards, seeds were washed in running water on a sieve (mesh 06, 23 BWG, rim 65 cm), until the mucilage was completely removed. Then, the seeds were left to dry in a dry, shaded, ventilated area at room temperature (25 °C±2). Before the laboratory tests, all seeds were disinfected after homogenization, following a method adapted from Silva et al. (2007). Then, they were immersed for 10 minutes in sodium hypochlorite solution (2.5% active chlorine) at a ratio of 10 mL per 1,000 mL water. Subsequently, they were rinsed in running water to fully remove the integument, and then left to dry on paper towels at ambient temperature (25 °C±2).

The second trial followed a fully randomized design, in a 4x2 factorial scheme [4 accessions (A1, A2, A3, and A4) and two fruit positions (proximal and distal), in which seeds were removed and treated with 500 mg L^{-1} gibberellic acid (GA_3)], with 4 replications of 25 seeds each.

The first counting of germinated seeds (FC) was done along with the germination test. For that, the number of seeds with primary root protrusion was computed on the 15th day after experiment implementation, with results expressed as a percentage (%) of germinated seeds. The germination test (G%) was conducted with four replicates of 25 seeds per treatment. To this test, the seeds were spread on Germitest papers moistened with distilled water (2.5 times its dry weight), which were placed into Gerbox boxes. Then, they were put in BOD chambers at constant temperature (25 °C±2), air relative humidity between 80-85%, and 12-h photoperiod (BRASIL, 2009). Evaluations were carried out from the 15th until the 105th day of the experiment setup (Silveira et al., 2019; Braga Filho et al., 2014). Germination (G%) was analyzed following the botanical criterion, in which only seeds with primary root protrusion are considered germinated. The results were expressed as a percentage (%).

Germination speed index (GSI) was also calculated in conjunction with the germination test, using daily values of germinated seeds. The calculations were made weekly from the first to the 105th day after sowing Maguire (1962). Moreover, mean germination time (MGT) was estimated so that the number of days required to reach maximum germination could be predicted Edmond e Drapala (1958).

For accessions, the quantitative-response variables for fruit characterization were subjected to a descriptive analysis to determine minimum, maximum, mean, and standard deviation values. Then, data on descriptive and germination analyses were submitted to normality analysis by Shapiro Wilk (p> 0.05). After analysis of variance (ANOVA), the means were compared by Tukey's test at 5% error probability, using the SISVAR statistical software.

Results and Discussion

Mean fruit transverse diameter (TD) was about 48.94 mm (40.90 mm minimum and 62.70 mm maximum), varying by 19.20 mm (Table 1). For this factor, the accessions showed significant differences and A3 had the smallest value (46.29 mm). Mean longitudinal diameter (LD) was 46.36 mm (31.10 mm minimum and 61.70 mm maximum), varying by 32.71 mm. For this parameter, A1 and A2 stood out as the best, with values of 51.88 and 48.30 mm, respectively, besides differing statistically from the others.

Barros et al. (2018) characterized fruits and seeds of araticum (*Annona crassiflora* Mart.) from the Cerrado of Mato Grosso State and found higher diameters (125.00 mm minimum and 163.50 mm maximum) when compared to this study. On the other hand, Braga Filho et al. (2014) observed significant variations in araticum fruit diameter both between collection areas and plants. Like other traits, there may be genetic variability for fruit diameter between araticum individuals. This is because the species has not yet been domesticated and presents a cross-pollination mechanism (Pimenta et al., 2014). For Ataíde et al. (2012), fruit length and diameter are major parameters for biometric studies since they determine fruit shape.

Mean fruit biomass (FB) was 63.41 g (29.00 g minimum and 130.40 g maximum), varying by 279.22 g (Table 1). The accessions showed significant differences, with A1 having the highest value (78.54 g) but not differentiating from A2 (67.40 g). Conversely, the lowest values were observed for A3 and A4 (55.74 g and 51.94 g, respectively).

Table 1. Descriptive statistics and average data of the physical characterization of the access fruits (A1, A2, A3 and A4) of araticum fruits.

Descriptive statistics of fruits							
	TD (mm)	LD (mm)	FB (g)	EB (g)	PF (g)	NS (un.)	TSM (g)
Average	48.94	46.36	63.41	22.78	39.63	38.0	286.3
Maximum	62.70	61.70	130.40	53.30	98.49	53	345.89
Minimum	40.90	31.10	29.00	12.90	6.09	11	205.18
Variation	19.20	32.61	279.22	65.24	185.91	58.7	12.45
DV.P	4.38	5.71	16.71	8.08	13.99	7.66	5.15
C.V. (%)	4.91	4. 12	11.78	14.97	15.45	5.77	4.95
Average data of the physical characterization of the fruits							
Acess	TD (mm)	LD (mm)	FB (g)	EB (g)	PF (g)	NS (un.)	TSM (g)
A1	52. 89 a*	51. 88 a	78.54 a	32.19 a	44.08 a	35 b	307.4 a
A2	48.09 ab	48. 30 a	67.40 ab	22.50 b	44.84 a	43 a	296.1 ab
A3	46.29 b	43. 55 b	55.74 b	18.25 b	36.72 a	40 a	283.9 b
A4	48.50 ab	41. 69 b	51.94 b	18.19 b	32.87 a	32 b	257.9 c

* Averages in the column followed by equal letters do not differ statistically by the Tukey test, at 5% probability of error. C.V. (%) = coefficient of variation. DV.P = standard deviation. A = accessions of araticum plants, TD = transversal diameter, LD = longitudinal diameter, FB = fruit biomass, EB = epicarp biomass, FP = fruit pulp, NS = number of seeds and TSM = mass of 1000 seeds.

According to Santos et al. (2011), fruit weight is a key trait both for the fresh fruit market and for the food processing industry. Heavier and uniformly-sized fruits are more attractive and improve concentrated pulp yield. Pereira et al. (2017) pointed out that fruits and seeds vary with mother-plant physiology in terms of weight and size, which is directly influenced by the relationship among soil, plant, and environment.

Mean epicarp biomass (EB) was 22.78 g (12.90 g minimum and 53.30 g maximum), varying by 65.24 g (Table 1). The accessions differed significantly in EB, and A1 (32.19 g) was superior to the others (Table 1). It is worth emphasizing that EB is an important parameter in post-harvest and has high genetic variability. According to Costa et al. (2015), the processing industry considers fruits with less peel and lower number of seeds to be of high yield. Dutra et al. (2017) reported that, while pulping fruits, part of the pulp remains adhered to the peel, which increases residue volume. Nonetheless, this factor was not evaluated in the present study.

Mean fruit pulp (FP) was 39.63 g (6.09 g minimum and 98.49 g maximum), varying by 185.91 g (Table 1). However, the accessions showed no significant differences for this trait. Braga Filho et al. (2014) reported that this is a fundamental parameter in araticum fruit evaluation, as the pulp is a part of economic importance (Gonçalves et al., 2013).

The number of seeds (NS) per fruit averaged 38 seeds (11 minimum and 53 maximum), varying by 58.7 (Table 1). For this parameter, A2 and A3 were the best accessions, with 43 and 40 seeds per fruit respectively, and differed statistically from the others. Interestingly, A1 had higher fruit biomass (78.52 g) but fewer seeds (35 seeds), whereas A3 had less fruit biomass (43.55 g) but more seeds per fruit (40 seeds).

Seed production can widely vary among accessions of a same species in a population, besides being directly influenced by biological factors of the plant (Dionisio et al., 2019). This is because each accession might have been cultivated in different soil, water availability, and microclimate conditions, which can directly interfere with its flowering and fruiting (Mendonça et al., 2014).

Regarding the NS, there are twofold aspects. For forest seedling production, fruits with greater amounts of seeds are more desirable. Whereas for the sale of fruits at both fresh and processed markets (juice, sweets, and ice cream), fewer seeds are most convenient since the pulp is the commercialized part (Gonçalves et al., 2013). Based on this, our study becomes important for showing the need to increase knowledge frontier on the subject.

Mean 1,000-seed mass (TSM) was 286.3 g (Table 1), which is proportionally less than that described for *A. crassiflora* of 639.4 g (Machado et al., 2016) and 681.17 g (Barros et al., 2018). Again, the accessions showed significant differences, with A1 having the highest value (307.4 g) but not differing from A2 (296.1 g); however, A3 and A4 showed the lowest values (283.9 and 257.9 g, respectively).

Seed physiological quality can be associated with TSM changes. Seeds of smaller sizes or lower TSM may be inferior in terms of physiological potential (Barbieri et al., 2013). However, such information is quite divergent in the literature, mainly for annual crop species. For these, several authors have claimed the lack of relationship between physiological attributes and seed size or TSM, but between these and specific mass or gravimetric weight.

According to Carvalho and Nakagawa (2012), seed weight provides information on seed quality and maturity and is directly influenced by moisture rate. In our study, this physical attribute of fruits was not determined or standardized.

The germination test showed that there was no interaction between accessions and seed position in the fruit. Neither was there isolated effects for these factors when considered parameters studied. Table 2 shows the absence of significant differences in FC, G%, GSI, and MGT means for araticum seeds.

Both FC and G% had high coefficients of variation (CV%) (Table 2), so the data were heterogeneous and may be related to seed dormancy. Similar results were found by Braga Filho et al. (2014), Barros et al. (2018), Silveira et al. (2019), and Pimenta et al. (2019). All these authors evaluated dormancy breaking in araticum seeds (*Annona crassiflora*).

Table 2. Average data of first germination count (FC), germination (G), germination speed index (GSI) and average germination time (MGT) of araticum seeds.

Acess	FC (%)	G (%)	GSI	MGT (days)
1	7 a*	20 a	0.24 a	26.73 a
2	8 a	17 a	0.21 a	23.43 a
3	8 a	21 a	0.26 a	22.27 a
4	9 a	16 a	0.20 a	26.29 a
C.V.(%)	48.63	39.82	7.86	16.62
Position of seed in the fruit	FC (%)	G (%)	GSI	MGT (days)
Proximal	9 a	20 a	0.25 a	23.82 a
Distal	7 a	16 a	0.20 a	23.54 a
C.V.(%)	48.63	39.82	7.86	16.62

*Averages in the column followed by equal letters do not differ statistically by the Tukey test, at 5% probability of error.

Seed position in the fruit had no relationship with its physiological quality, corroborating literature findings. Some studies such as that of Dionisio et al. (2019) with Brazil nut (*Bertholletia excelsa*), that of Lessa et al. (2014) with tamboril (*Enterolobium contortisiliquum*), and that of Freitas et al. (2013) with sabiá (*Mimosa caesalpiniifolia*) seeds have shown that their locations in the fruit does not influence the germinative performance or seedling formation. By contrast, for catingueira seeds (*Poincianella pyramidalis*), Mendonça et al. (2016) found that seed position in the fruit had influence on germination. These authors stated that, despite having a shorter length, seeds in a proximal position show higher germination rates.

In the present study, we evidenced that seed position in the fruit does not interfere with germination for the studied species. Therefore, from an economic and technical point of view, we do not recommend classifying seeds by their position in the fruit. As there is little information regarding the effect of this parameter on germination in different plant species, studies should be developed to improve seed market security and the production of quality seedlings.

It is also important to highlight that, from a physiological point of view, all traits measured here can be influenced by the productive environment and genetic load of individuals. Despite these, our findings are vital to improving the knowledge of this species to be used in future breeding programs for the commercial exploitation of araticum.

Conclusions

Araticum fruits had mean biomass of 63.41 g, a mean number of 38 seeds per fruit, and a mean pulp weight of 39.63 g.

Araticum seed germination was not influenced by accessions or seed position in the fruit.

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