

Morphological characterization of fruits, seeds and seedlings of araticum plant (*Annona crassiflora* Mart – Annonaceae)¹

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ABSTRACT – Araticum (*Annona crassiflora* Mart. - Annonaceae) is a species with great ecological importance and potential for fruit production. Its fruits, seeds and seedlings were morphologically described in this article as being the main objective of this study. To describe and illustrate the examples, 100 seeds and 42 fruits were analyzed; the latter being collected in Santo Antônio do Leverger - MT - Brazil, in 2011. To describe the morphology of the seedling, 100 seeds were germinated in two different environments. The araticum plant has a compound, multiple strobiliform, globose-subglobose and fleshy fruit, with average measures of 12.9 cm length, 13.5 cm width, 12.7 cm thickness, weight of 1,187.0 g and 99.2 seeds per fruit. The seeds are obovoid, hairless, smooth with a bony aspect and a pale brown color; they have an average of 17.7 mm length, 10.8 mm width and 8.3 thickness. Epigeous and phanerocotylar germination begins about 148 days after sowing. All morphological data of araticum fruits, seeds and seedlings can be used to recognize the botanical family and, when associated to other features, to recognize the species in the field.

Index terms: Annonaceae, araticum, marolo, morphology.

Caracterização morfológica de frutos, sementes e plântulas de araticunzeiro (*Annona crassiflora* Mart – Annonaceae)

RESUMO - Objetivou-se com o presente estudo descrever a morfologia de frutos, sementes e plântulas de araticunzeiro (*Annona crassiflora* Mart. - Annonaceae), uma espécie com alto potencial frutícola e grande valor ecológico. A caracterização das estruturas foi realizada a partir de 42 frutos e de 100 sementes, coletados no município de Santo Antônio do Leverger - MT, em 2011. Para descrever a morfologia da plântula, 100 sementes foram colocadas para germinar, em dois ambientes distintos. O araticunzeiro apresenta fruto composto, múltiplo estrobiliforme, globoso-subgloboso e carnoso, com médias de 12,9 cm de comprimento, 13,5 cm de largura, 12,7 cm de espessura, massa de 1.187,0 g e de 99,2 sementes por fruto. As sementes são obovoides com tegumento glabro, castanho-claro, opaco, liso e ósseo; possuem em média, 17,7 mm de comprimento, 10,8 mm de largura e 8,3 mm de espessura. A germinação é epígea e fanerocotiledonar e se inicia após 148 dias da semeadura. Os aspectos morfológicos do fruto, da semente e da plântula de araticunzeiro podem ser utilizados para o reconhecimento da família botânica e, se associados a outras características, para o reconhecimento da espécie no campo.

Termos para indexação: Annonaceae, araticum, marolo, morfologia.

Introduction

The Annonaceae family comprises 130 genera and 2,200 species, distributed predominantly tropically. In Brazil there are 33 genera that are home to about 250 species (Souza and Lorenzi, 2005), including wild and cultivated, many of which have high-quality fruits, especially those belonging to the genus *Annona* (Joly, 1998).

The araticum tree (*Annona crassiflora* Mart. - Annonaceae), also known as Marolo or Araticum do cerrado, is native to the Brazilian Cerrado, in the states of São Paulo, Mato Grosso do Sul, Mato Grosso, Minas Gerais, Goiás, Bahia and Tocantins (Lorenzi, 2002). It is an arboreal plant, which can reach up to 8 m of height (Silva et al., 2001; Lorenzi, 2002), whose winding stem, 20-30 cm in diameter, is covered with a rough and very thick bark, which provides resistance to

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the action of fire (Lorenzi, 2002). The araticum tree leaves are alternate, distichous, simple and without stipules (Souza and Lorenzi, 2005; Lobão et al., 2005) and the flowers are isolated, axillary, actinomorphic with three sepals, six petals and numerous stamens and carpels (Lobão et al., 2005). The fruit is a multiple strobiliform type, glabrous, of subglobose form, composed of numerous small fruits that mostly contains a single seed. The tubercular and papillose epicarp staining is green when immature and greenish brown when mature (Lima-Brito et al., 2006).

The araticum tree is one of the most frequent species in the Cerrado biome, exploited by the local population for food and medicinal use. By presenting appreciable aroma, flavor and digestibility, the pulp of its fruit is consumed fresh or processed for the production of juices, liqueurs, ice cream and jellies (Telles et al., 2003). Besides the organoleptic characteristics, the fruit has a high nutritional value, with significant levels of lipids, fibers, calories, magnesium, phosphorus and antioxidants (Damiani et al., 2011). Furthermore, the araticum tree can also have an allelopathic effect on other species, inhibiting germination and/or seedling development of Marandu grass (*Brachiaria brizantha* Stapf.) and of soybean (Inoue et al., 2009), plus an insecticide effect, which enables its use in controlling the bug vector of dengue fever (*Aedes aegypti* L.) (Omena et al., 2007) and of brown stink bug (*Euschistus heros* Fabr.) (Oliveira and Pereira, 2009).

The study of fruits, seeds and seedlings, besides contributing to the knowledge of the species, generates information necessary for taxonomic and phylogenetic purposes, adding to the traditional analyzes of vegetative and floral organs (Oliveira, 2001). Thus, morphological studies assist in interpreting laboratory tests and allow the identification of botanical species, constituting an important tool to recognize it in the seed banks in the soil and in seedling stage in forest formations (Melo et al., 2004). The knowledge of the morphology of native plants is of great value to understanding the behavior of the species in relation to the mechanisms of dispersal, succession, natural regeneration and conservation (Melo et al., 2004; Cosmo et al., 2010).

The use of morphological characters with taxonomic purpose is especially important for Annonaceae, considering that this family is one of the most uniform, both in terms of anatomical and structural points of views, of habit and habitat, with a combination of striking and peculiar characters (Barroso et al., 1978). This information is confirmed by the morphology of the ruminated seed and tiny embryo, which constitutes a good character of identification at the family level, although the identification of some genera is

hampered by the lack of data regarding the fruits and they are underrepresented in spaces for collection of information about fruit trees and fruits and herbaria (Barroso et al., 1999).

The morphology of fruits, seeds and seedlings of the araticum tree was studied by Lima-Brito et al. (2006), from a native population in the Brazilian State of Bahia. However, similar studies are necessary with populations of different regions, because phenotypic variations in response to the heterogeneity are expected in the cerrado plants, as reported by Rocha Filho and Lomônaco (2006).

Accordingly, the aim of the present study was to describe and illustrate the morphology of fruits, seeds and seedlings of *Annona crassiflora* Mart.

Materials and Methods

Fruit collection - fruits were collected from February to March 2011, from mother plants selected in the araticum tree native population belonging to the Cascata do Andorinha farm, located in the geographic coordinates 15° 50' S and 55° 21' W, at 770 m of altitude, in the municipality of Santo Antônio do Leverger (MT). For species identification, samples were collected from the plant material and compared with the exsiccated specimen registered under number 23,134, deposited in Herbário Central at Universidade Federal de Mato Grosso, in Cuiabá (MT). Forty two fruits were collected immediately after their dispersion from 17 mother plants, being collected randomly, in order of maturity. After collection, the fruits were packed in polypropylene trays (50 x 31 x 8 cm) and transported to Instituto Federal de Educação, Ciência e Tecnologia de Mato Grosso, *Campus São Vicente* (IFMT *Campus São Vicente*), located in the municipality of Santo Antônio do Leverger (MT), where the assessments related to the morphological characterization of the fruits were conducted.

Morphological characterization of the fruits - to determine the length, width and thickness of the fruit was used a digital caliper graduated from 30 cm and accuracy of 0.001 cm, positioned at the apex at the scar of the stalk to length, the width being the largest transverse axis and the thickness the smallest transverse axis. To measure the mass of the fruit, was used a digital scale with a capacity of six kilograms (13 pounds), accurate to 0.002 g and the number of seeds per fruit was verified after manual pulping. In addition to the measurable variables, were observed in the fruit: type, shape, texture, surface and color, performing transverse and longitudinal sections in the pericarp for observation of detail when necessary.

Seed processing and samples preparation: after pulping

the fruits, the seeds were washed in running water until complete removal of the pulp and dried in the shade for 48 hours on paper towel. The seeds with malformation, cracks or holes in the seed coat were removed. Whole seeds were packed in "Kraft" paper bag and transported to Laboratório de Análise e Tecnologia de Sementes of the Departamento de Fitotecnia e Fitossanitarismo of the Universidade Federal do Paraná, in Curitiba (PR). Subsequently, the average sample was homogenized and divided into duplicate sample and working sample, using a centrifugal divider. Both samples were kept in a dry chamber at a temperature of 17 ± 2 °C and relative humidity of 60-70%, until being used.

Morphological characterization of the seeds: in April 2011 were randomly drawn 100 seeds from the working sample for morphological description and illustration. The dimensions of length, width and thickness were measured with a digital caliper graduated 20 cm and accuracy of 0.001 g.

In seeds were externally observed: shape, color, texture, consistency, presence of aryl and positions of hilum and micropyle. For internal descriptions regarding the presence of endosperm, type, position and shape of the embryo and the position of the radicle-hypocotyl axis, transverse and longitudinal sections were performed in the seed, using a scalpel blade and observation in stereoscopic microscopic (Abreu et al., 2005; Rego et al., 2010). For better observation of the embryo, 60 seeds were imbibed in distilled water for 24 hours at 25 °C, followed by removal of the integument and longitudinal section parallel to the embryonic axis. Subsequently, was performed staining of the embryos on filter paper moistened in 2.5 times the mass of the paper, with a tetrazolium solution (2,3,5 triphenyl tetrazolium chloride) 0.5% for six hours in an incubator with a temperature of 25 °C.

Morphological characterization of the seedlings: 100 seeds were put to germinate in polypropylene boxes containing vermiculite as substrate, moistened in its field capacity. The boxes were maintained in an incubator without controlling environmental conditions. 100 seeds were also sown on paper

towel roll, moistened with water at a ratio of 2.5 times its dry weight. The rolls were kept in an Mangelsdorf type incubator, at 25 °C and constant light of 110 Lx. The phases between the swelling of the endosperm and the full development of protophylls were observed. To characterize the seedling were observed type, shape, color and hairiness of the root and of the hypocotyl and consistency, texture, shape, color, veining and type of maple, apex and base of the protophylls. The technological procedures for the study of the morphology of the fruit, seed and seedling were based on the work by Barroso et al. (1999).

Statistical procedures – the data of the quantitative variables were electronically tabulated and analyzed, determining the minimum, average and maximum values with their respective standard deviation and coefficient of variation.

Results and Discussion

Morphological characterization of the fruits – the araticum tree fruits (Figure 1) are of the composite type, multiple strobiliform, according to the classification from Barroso et al. (1999), globose to subglobose in shape and with a fleshy consistency. 33-152 seeds per fruit were found. The fruits are, on average, 12.9 cm long, 13.5 cm wide, 12.7 cm thick and 1,187 g (0.00261688705 pounds) weight (Table 1). The fruits are formed by many monospermic small fruits (Figure 1C), but occasionally in some were found two or more seeds, which had a modified shape over the species standard. The fruits are composed by mesocarp and endocarp, fleshy consistency, sincarpic, coalescing, non drupoid and arranged on a fleshy and taper receptacle (stalk) (Figure 1D). The epicarp is glabrous, tubercular and papillose (Figures 1A, 1B, 1C, 1D) and staining is green when immature and greenish brown when mature.

The morphological information of the fruit of the araticum tree agree with the description of Lima-Brito et al., (2006), in a study of the morphology of fruits of the Annonaceae family, including *Annona crassiflora*.

Table 1. Length, width, thickness, fresh mass and number of

seeds/fruit of fruits of *Annona crassiflora*.

Variable	Minimum	Average	Maximum	Standard deviation	Coefficient of variation (%)
Length (cm)	9.5	12.9	18.0	1.9	14.4
Width (cm)	10.5	13.5	17.8	1.5	10.8
Thickness (cm)	9.9	12.7	16.6	1.5	11.6
Fresh mass (g)	515.0	1,187.0	2,040.0	365.9	30.8
Number of seeds/fruit	33.0	99.2	152.0	33.4	33.7

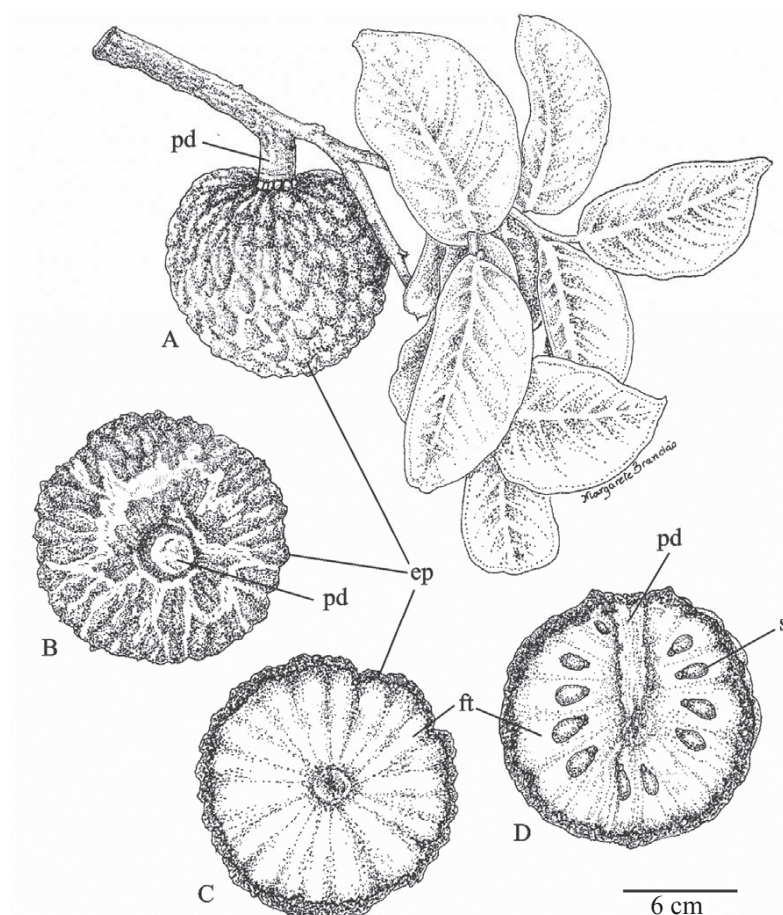


Figure 1. Fruit of *Annona crassiflora*. (A) branch with fruit and external appearance in lateral view; (B) external appearance in basal view; (C) internal appearance in cross-sectional view; (D) internal appearance in longitudinal view. pd: stalk; ep: epicarp; ft: small fruit; s: seed.

The fruit mass of the araticum tree obtained in this work is comprised within the range of 0.5 to 4.5 kg per fruit mentioned by Silva et al. (2001), however, with average mass higher than those found by Lima-Brito et al. (2006) found an average of 836.3 g in fruits of the araticum tree collected in the Brazilian state of Bahia. The values for fruit mass found in the present work, as well as those cited by other authors, emphasize the importance of the species, as a fruit plant, because the size of the fruit greatly surpasses other species of the genus that are already grown for this purpose and whose fruits have wide acceptance in consumer markets, particularly in their centers of origin. Among them it is possible to mention sugar-apples (*Annona squamosa* L.) and custard-apple (*Annona cherimola* Mill. x *Annona squamosa* L.), with average masses of 231 g (Lima-Brito et al., 2006) and 275 g to 368 g (Neves and Yuhara, 2003), respectively. Regarding the size of the fruit, the araticum tree can be compared to Brazilian pawpaw (soursop, prickly custard apple, Soursapi) (*Annona muricata* L.), which has a mass of approximately 1 kg (Lima-Brito

et al., 2006) and with wild sugar apple or aratiku (*Rollinia mucosa* (Jacq.) Baill.) which produces fruits of mass equal to 1.3 kg (Lorenzi, 2002).

Morphological characterization of the seeds – the seeds of the araticum tree are obovoid (Figure 2A) and in average are 17.7 mm long, 10.8 mm wide and 8.3 mm thick (Table 2). The integument is glabrous, light brown in color, opaque, with a smooth texture and a bony consistency. The tegmen is comprised of fibrous layers that fall within the endosperm resulting in ruminations (Figure 2E), as described by (Lima-Brito et al., 2006). It was observed that after seed drying, the tegmen fibers detach and the assembly formed by the endosperm and embryo becomes loose within the integument. The araticum tree seeds have basal aryl of rough surface and irregular and conspicuous boundary (Figures 2A, 2B, 2C and 2D) arranged around the hilum and the micropyle. The hilum is basal, miniature and irregularly shaped between circular and oval (Figure 2C) and closed by a fold of buffer, corroborating the information

cited by Barroso et. al. (1999) for the Annonaceae family. After removal of the aryl it was possible to see the micropyle (Figure 2D), forth below and in continuity with the hilum. The endosperm is ruminated, thick, abundant (Figure 2F) and whitish-yellow in color. The embryo is basal and crude,

approximately 2 mm long (Figures 2F, 2G and 2H), hyaline and gelatinous. It was observed in the embryonic axis the region of the radicle and cotyledons (Figure 2H), although for most seeds, these structures had not fully differentiated, similar to the observations by Silva et al. (2007).

Table 2. Length, width and thickness of seeds of *Annona crassiflora*.

Variable	Minimum	Average	Maximum	Standard deviation	Coefficient of variation (%)
Length (mm)	13.4	17.7	22.7	2.3	13.1
Width (mm)	9.0	10.8	13.6	0.9	8.7
Thickness (mm)	6.4	8.3	11.2	0.8	9.7

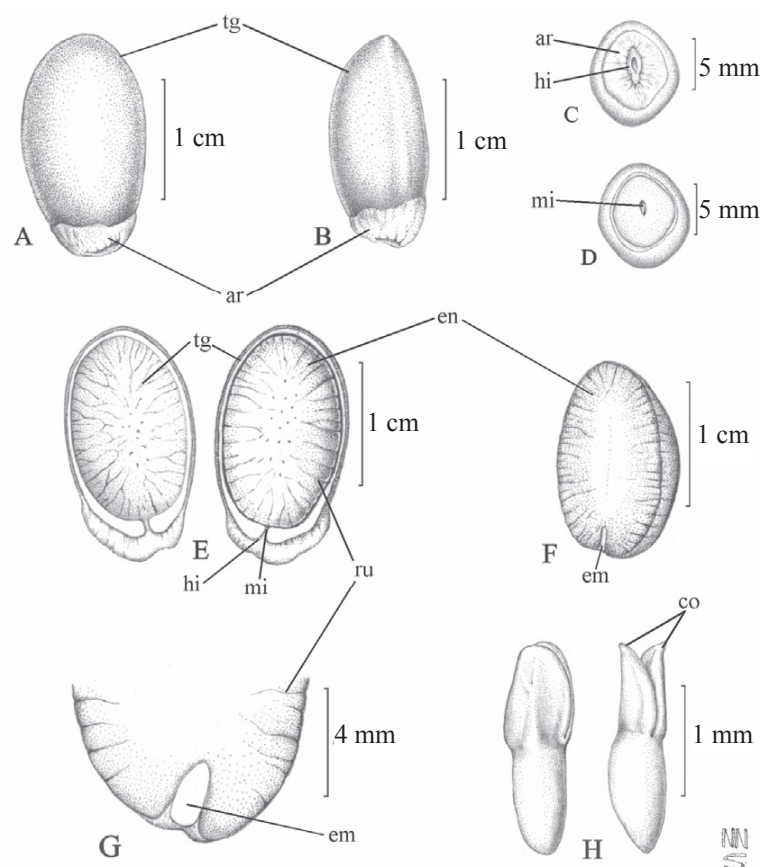


Figure 2. Seed of *Annona crassiflora*. (A) lateral view; (B) dorsal view; (C) basal view; (D) basal view without the aryl; (E) integument and endosperm; (F) longitudinal section of the endosperm; (G and H) embryo. tg: integument; ar: aryl; mi: micropyle; hi: hilum; en: endosperm; ru: ruminated; em: embryo; co: cotyledon.

The morphological characteristics of the seed found in this experiment are equivalent to the information about the species cited by Lima-Brito et al. (2006) and Silva et al. (2007).

Among the features described for the seed, the embryo size and ruminations are of fundamental importance for the botanical identification of individuals at family level because they are specific conditions of Annonaceae (Barroso et al., 1999).

According to Barroso et al. (1999), the embryo is small because it develops very late after the seed is almost entirely formed, which becomes an ecological mechanism for survival of the species, either for the time required for germination (Silva et al., 2007), which enables the new individual a more favorable environment for their development and/or the formation of persistent seed banks in the soil (Cavalcante et al., 2007).

Morphological characterization of the seedlings – after 148 days of sowing was observed a longitudinal rupture of the integument caused by swelling of the endosperm (Figure 3A) and primary root protrusion (Figure 3B), followed by elongation of the primary root and hypocotyl (Figure 3C), secondary root formation (Figure 3D), expansion of cotyledons parallel to the development of protophylls (Figure 3E) and ending with the seedling formed (Figure 3F), in approximately 190 days after sowing. The long period required for seed germination and seedling development of this experiment is due to the morphophysiological numbness peculiar to the species (Silva

et al., 2007) and most likely to the low ambient temperatures recorded during the test.

The germination of the araticum tree is epigeal and phanerocotylar, which starts by primary root protrusion in the region of the micropyle (Figure 3B). The primary root is axial, cylindrical, of pale cream-white color, glabrous and long, with secondary roots originating from the base of the collect, which resemble the main root, but thinner and shorter; the hypocotyl is fleshy, thick, glabrous and white in color and/or slightly pink, which holds at its apex the endosperm and other overhead structures still surrounded by the integument (Figure 3D).

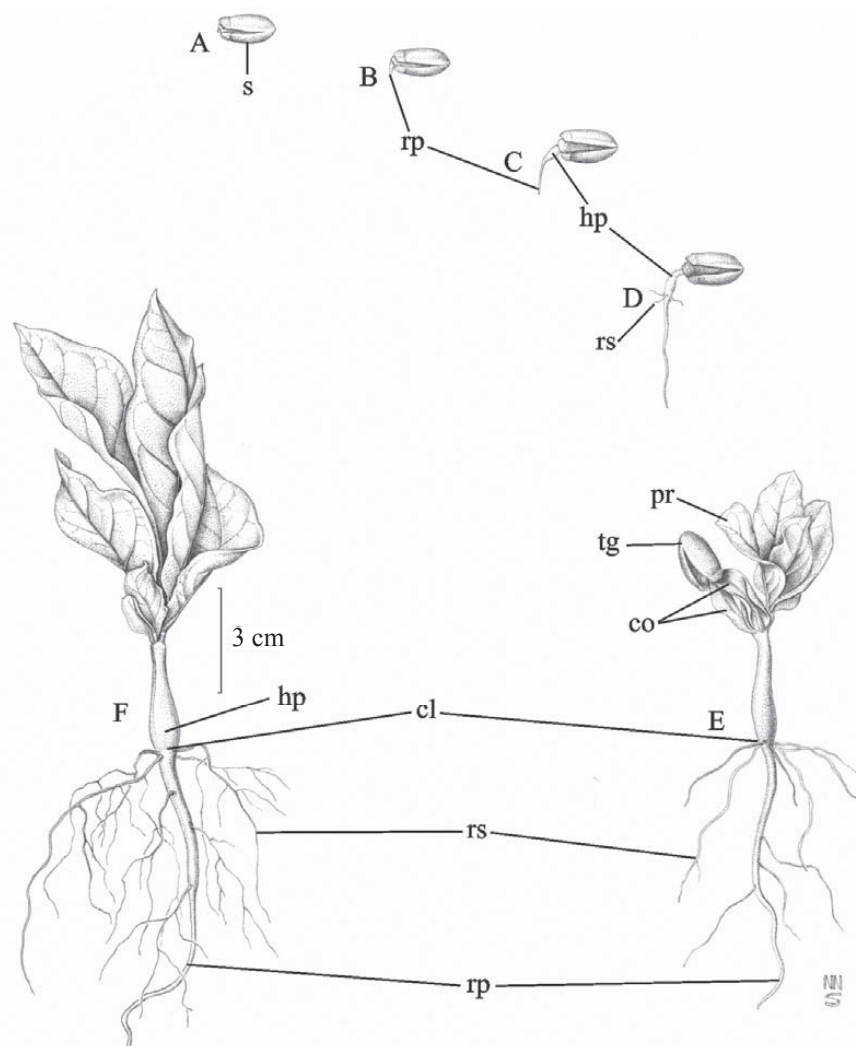


Figure 3. Germination of *Annona crassiflora*. (A – D) initial phase; (E – F) details of the seedling. s: seed; rp: primary root; hp: hypocotyl; rs: secondary root; pr: protophyll; tg: integument; co: cotyledons; cl: collect.

Consumption of the araticum tree seeds reserves is slow, and even after the expansion of the cotyledon leaves and the development of the first leaves (protophylls), it is possible to observe residues of the endosperm and the integument still adhering to the

seedling (Figure 3E). When the seedlings are formed it is observed a complex root system formed by the primary and thick root close to the collect and tapering towards the apex, and from it and from the base of the collect emerge secondary roots, which branch

out and complete the root system (Figure 3F). The hypocotyl is thick, with a thicker base with respect to the apex, in which the fully developed cotyledons and protophylls are observed. The cotyledons and protophylls are petiolate and innervated, and the protophylls and adult leaves have similar characteristics as the limbus and the ribs. Leaves are simple, elliptic, glabrous of obtuse to slightly acute apex and wedge-shaped base.

Conclusions

The araticum plant has a compound, multiple strobiliform, globose-subglobose and fleshy fruit, with average measures of 12.9 cm length, 13.5 cm width, 12.7 cm thickness, weight of 1,187.0 g and 99.2 seeds per fruit. The seeds are obovoid, hairless, smooth with a bony aspect and a pale brown color; they have an average of 17.7 mm length, 10.8 mm width and 8.3 thickness.

Epigeous and phanerocotylar germination begins about 148 days after sowing.

All morphological data of the araticum fruits, seeds and seedlings can be used to recognize the botanical family and, when associated to other features, to recognize the species in the field, provided that associated with other characteristics, such as those observed in leaves and flowers.

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