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ORIGINAL ARTICLE

Anatomy of stems, leaves, roots and the embryo of *Garcinia brasiliensis* Mart. - Clusiaceae

Anatomia de caule, folhas, raízes e do embrião de Garcinia brasiliensis Mart. - Clusiaceae

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ABSTRACT: Studies concerning the structure of plant embryos are very important in different areas of study such as plant systematics and evolution or the handling of native species. This work aims at evaluating the anatomy of the embryo and vegetative organs of *Garcinia brasiliensis* in order contribute to taxonomical, ecological, handling studies of this species. Some seeds were stored for anatomical analysis of the embryo and some were germinated to obtain young plants for anatomical analysis of the vegetative organs. Plants were cultivated in growth chamber for 120 d. Seeds and young plants were both fixed in a solution of formaldehyde, acetic acid, and 70% ethanol and usual plant microtechnique methods were employed. Slides were photomicrographed and a quantitative analysis of tissues was performed using UTHSCSA-Imagetool software. Histochemical analysis was performed for the detection of starch, lipids, alkaloids, and phenolic compounds. The *G. brasiliensis* embryo has rudimentary cotyledons and the reserves are stored in the hypocotyl-radicle axis. Most of the reserves are starch grains, stored in ground meristem cells, which contain a few oleossomes and alkaloids in the cell walls. The young plant leaves show dorsiventral and hypostomatous properties, including a one-seriated epidermis and a thick cuticle. In the midrib vascular bundle, the phloem is organized around the xylem vessels. The primary stem has a one-seriated epidermis and angular collenchyma. Adventitious roots are polyarch and show a one-seriated epidermis and endodermis. The anatomy of embryos and young plants of *G. brasiliensis* is similar to that of other Clusiaceae species.

RESUMO: *Estudos sobre a estrutura do embrião são de enorme importância para diferentes áreas, como a sistemática, a evolução e o manejo de espécies nativas. Portanto, o objetivo deste trabalho foi avaliar a anatomia do embrião e de órgãos vegetativos das plântulas de Garcinia brasiliensis. Sementes foram reservadas para realização das análises anatômicas do embrião e outras foram postas para germinar, para obtenção das plantas visando à análise de folhas, caules e raízes. As plantas foram cultivadas em sala de crescimento por 120 dias. Sementes e plantas foram fixadas em uma solucion de formaldeído, ácido acético e etanol a 70% e submetidas a procedimentos usuais de microtécnica vegetal. As lâminas foram fotomicrografadas e a análise quantitativa dos tecidos foi realizada no software UTHSCSA-Imagetool. Análises histoquímicas foram realizadas para detecção de amido, lipídeos, alcalóides e compostos fenólicos. O embrião de G. brasiliensis possui cotilédones pouco desenvolvidos, sendo a reserva das sementes localizada no eixo hipocótilo-radicular. As reservas são compostas principalmente de amido, localizado nas células do meristema fundamental. Há um pequeno número de oleossomos nas células do meristema fundamental e suas paredes celulares contêm alcalóides. A folha das plantas é dorsiventral e hipoestomática, com epiderme unisseriada e cutícula espessa. No feixe vascular da nervura central, o floema está localizado em torno do xilema. O caule em estádio primário de crescimento possui epiderme unisseriada e colênquima angular. As raízes adventícias são poliarcas, com epiderme unisseriada. A anatomia do embrião e das plantas jovens de G. brasiliensis é similar a outras espécies de Clusiaceae.*

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1 Introduction

The Clusiaceae family comprises seven genera and 1,100 plant species that are distributed in tropical and subtropical regions (STEVENS, 2007). *Garcinia* is the larger genus in Clusiaceae, containing 300 species, many of then found in Brazil (SACRAMENTO et al., 2007; ABDULLAH; ISMAIL, 2010; HEMSHEKHAR et al., 2011). One important Brazilian species is *Garcinia brasiliensis* Mart. called "bacupari", which is an Amazonian tree species, but is widely cultivated in Brazil for medical uses and fruit production (ZAMITH; SCARANO, 2004; LUCAS, 2008).

The epicarp of *Garcinia* fruits is hard and strong, often showing yellow or orange colors. However, the white and creamy mesocarp is edible (FRANCO et al., 2007). Bacupari fruits are classified as "bacoids" containing two or three seeds that are dispersed by animals (zoochoric) or by water (hydrochoric) (GALETTI et al., 2008; LUCAS, 2008). For *G. brasiliensis* of the Brazilian Savanah (Cerrado), only zoochoric dispersal has been reported (DIAS NETO et al., 2009).

Anatomical studies are important for solving taxonomical questions in Clusiaceae. For example, in *Clusia*, seeds are exalbuminous and the embryo is hypocotilar, showing very small cotyledons (MOURÃO; BELTRATI, 2001; PATHIRANA; HERAT, 2004; CAMPANA; MOURÃO; MARZINEK, 2010). According to Campana, Mourão and Marzinek (2010), the anatomies of the vegetative organs and embryo are both important for the circumscription of subfamilies and tribes among Clusiaceae. Likewise, anatomical and morphological studies are very important for solving the evolutionary questions, as well as establishing the relationships among *Garcinia* species (PATHIRANA; HERAT, 2004).

Despite the importance of anatomical studies in the Clusiaceae family, there have still been few studies of the anatomies of these species. Mourão and Marzinek (2009) reported the importance of anatomy for the *Garcinia* phylogeny but there is a lack of information for a large number of species. As there is no information in the literature about *G. brasiliensis* anatomy, the goal of this work is to evaluate the anatomy of the embryo, stem, leaves, and roots of *G. brasiliensis*.

2 Materials and Methods

The work was carried out at the Laboratório de Biotecnologia Ambiental & Genotoxicidade at the Universidade Federal de Alfenas (Alfenas, MG) and at the Laboratório de Anatomia Vegetal of Universidade Federal de Lavras (Lavras, MG). The plant material consisted of *G. brasiliensis* seeds harvested from a crop population cultivated in the Universidade Federal de Viçosa (Viçosa, MG). Seeds were obtained from different plants and were utilized for anatomical analysis of the embryo. Seeds were also used to the production of the plants used for anatomical analysis of stems, leaves, and roots.

Seeds were germinated in plastic trays containing vermiculite saturated with distilled water. Seeds were maintained in a growth chamber with continuous light with 84 μmol m^{-1} s^{-1} of photosynthetic photon flux density at 25 °C for 120 d. After this period of time, the stems, leaves, and roots were collected and fixed in a solution of formaldehyde, acetic acid, and 70% ethanol ($F.A.A_{70\%}$) for 72 h and then stored in 70% ethanol until further analysis (KRAUS; ARDUIN, 1997). The seeds were also fixed in $F.A.A_{70\%}$ for 72 h and then stored in 70% ethanol until further analysis (KRAUS; ARDUIN, 1997). Embryos were separated from the seed coats and investigations of the transversal sections at three different regions were conducted: the stem pole (0.5 cm from the stem apex), the median region, and the radicle pole (0.5 cm from the root apex). Next, the sections were cleared with 50% sodium hypochlorite, rinsed in distilled water twice for 10 min, stained with safrablau solution (1% safranin and 0.1% astra blue in a 7:3 ratio), and mounted on slides with coverslips with 50% glycerol (KRAUS; ARDUIN, 1997).

The slides were photographed under a Zeiss MicroImaging GmbH Scope.A1 (Carl Zeiss MicroImaging GmbH, Göttingen, Germany) coupled to a Canon A630 digital camera and the images were analyzed using UTHSCSA-Imagetool software (The University of Texas Health Science Center, San Antonio, Texas, USA). The anatomical characteristics evaluated were: cuticle thickness, secretory channel diameter, diameter of epithelium cells in secretory channels, xylem vessel diameter, phloem thickness, starch grain diameter, and diameter of the ground meristem cells. The experimental design was completely randomized with three treatments and five replicates. Three sections and one field were photographed for each replicate. The data were tested with one-way ANOVA and the Scott-Knott test to $p < 0.05$.

Histochemical analysis was performed in fresh (not fixed) embryos for the detection of starch grains (Lugol), lipids (SUDAN III), alcaloids (Draggendorf), and phenols (ferric chloride test), according to Kraus and Arduin (1997).

Anatomical analysis was performed in stem internodes, the median region of fully expanded leaves, and in the piliferous zone of roots from plants that were 120 d old. The fragments of stems, leaves, and roots were fixed in an $F.A.A_{70\%}$ solution for 72 h and then stored in 70% ethanol until further analysis (KRAUS; ARDUIN, 1997). Paradermal leaf sections were obtained using steel blades on their abaxial sides. Next, the sections were cleared with 50% sodium hypochlorite, rinsed in distilled water twice for 10 min, stained with 1% safranin solution, and mounted on slides with coverslips with 50% glycerol. The 2 cm leaf fragments taken from the midrib and stem internodes and roots fragment 2 cm away from the root apex were used for the cross sections using an LPC table microtome. Sections were cleared with sodium hypochlorite, rinsed in distilled water twice for 10 min, stained with safrablau solution, and mounted on slides with coverslips with 50% glycerol. The slides were photographed under a Zeiss MicroImaging GmbH Scope.A1 (Carl Zeiss MicroImaging GmbH, Göttingen, Germany) coupled to a Canon A630 digital camera.

The UTHSCSA-Imagetool software was used for image analysis and the following parameters were measured: polar diameter of the stomata, equatorial diameter of the stomata, stomatal density (stomata mm^{-2}), the stomatal ratio between polar/equatorial diameters, epidermal thickness of roots, stems, and the abaxial and adaxial sides of leaves, mesophyll thickness, palisade parenchyma thickness, spongy parenchyma thickness, cortex thickness of roots and stems, xylem vessel element diameter, and phloem thickness. Five replicates, three

sections for each replicate, and one field for each section were evaluated for each anatomical characteristic.

3 Results

G. brasiliensis embryos show rudimentary cotyledons (Figure 1A) and a one-seriated protodermis, which contains table-shaped cells and a thick cuticle (Figure 1B). However, the cuticle is thicker in the median region of the embryo (Table 1). The ground meristem is localized above the protodermis, showing round cells and secretory channels (Figure 1D). The epithelial cells and lumen diameter of the secretory channels are larger in the median region of the embryo (Table 1). The ground meristem cells are rich in starch grains and may show small oleosomes (Figure 1C). Ground meristem cells have equal diameters along the embryo axis (Table 1), as well as the same diameter as starch grains (Table 1). There are deposits of alkaloids in the ground meristem cell walls along the embryo

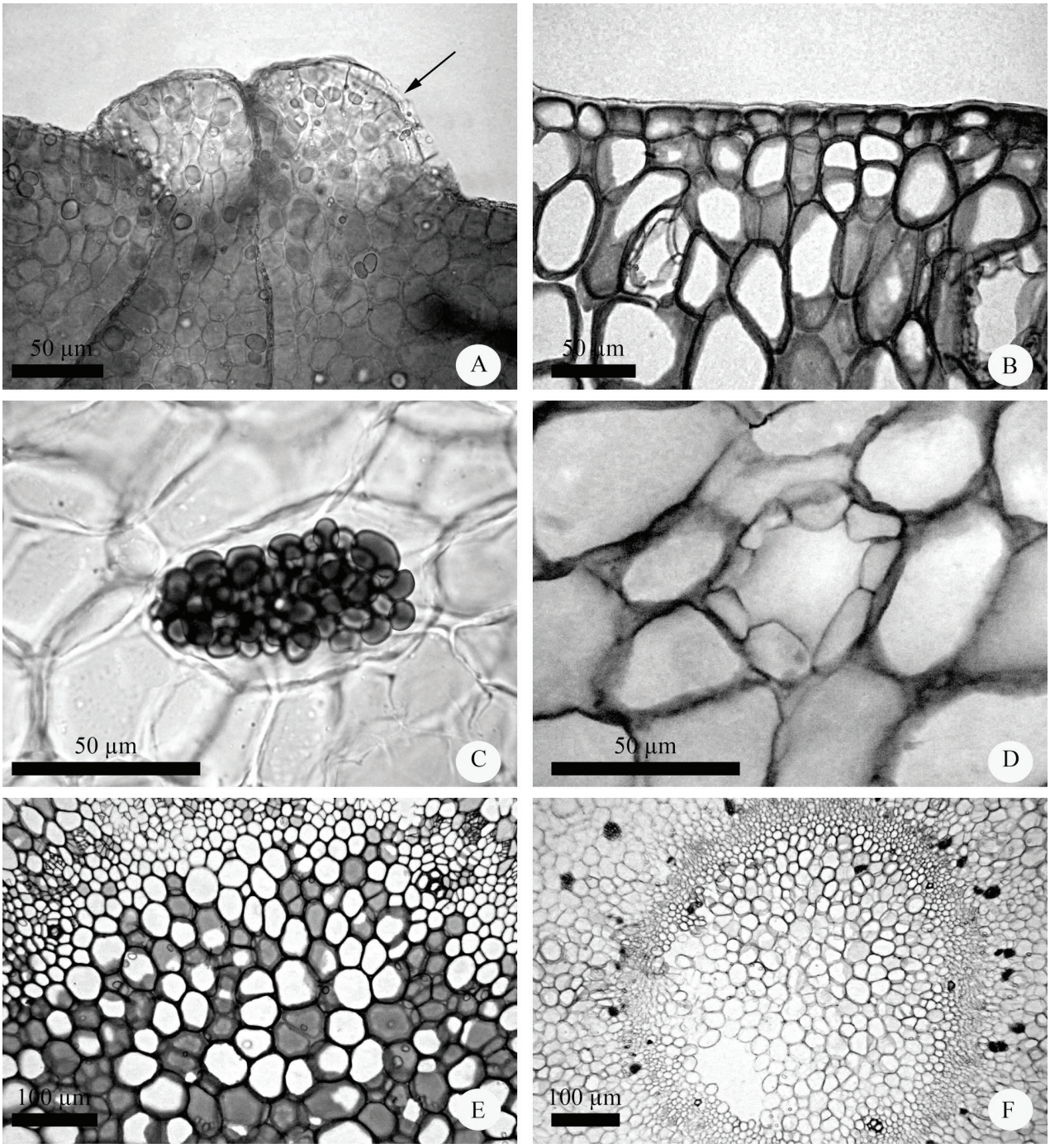

Figure 1. Transversal sections of the embryo of *Garcinia brasiliensis* showing details of the internal structure. A= stem pole showing rudimentary cotyledons (arrow), B= hypocotyl showing protodermis, cuticle, and ground meristem, which contain secretory channels, C= starch grains in the ground meristem cells of the hypocotyl-radicle axis, D= secretory channel in the ground meristem of the hypocotyl-radicle axis, E-F= procambium and pith showing ground meristem cells. Bars: A-D= 50 µm and E-F= 100 µm.

Table 1. Anatomical characteristics of the embryo of *G. brasiliensis* at different positions.

Characteristics	Stem pole	Median region	Root pole
Secretory channel lumen diameter (µm)	90.64 b	111.13 a	91.31 b
Secretory channel cells diameter (µm)	11.07 b	014.34 a	17.27 a
Cuticle thickness (µm)	03.73 b	004.56 a	03.76 b
Xylem tracheary element diameter (µm)	14.52 b	020.41 a	22.10 a
Phloem thickness (µm)	31.07 b	040.24 a	47.80 a
Parenchyma cell diameter (µm)	68.34 a	065.32 a	70.70 a
Starch grain diameter (µm)	09.87 a	010.31 a	10.90 a

Means followed by the same letters in the columns do not differ according to the Scott-Knott test ($p < 0.05$).

axis of *G. brasiliensis*, despite phenolic compounds not being detected.

Transversal sections reveal the procambium to be located in the center of the embryo axis (Figures 1E, F), which contains partially differentiated xylem vessels and phloem elements. Xylem element diameter and phloem thickness show larger values in the root pole and median region of the embryo (Table 1).

The leaves of *G. brasiliensis* reveal a dorsiventral structure containing palisade parenchyma at the adaxial side showing an average thickness of 41.66±3.47 µm. Spongy parenchyma is located on the abaxial side, with an average thickness of 191.83±16.43 µm. The leaf epidermis on both the adaxial and abaxial sides is one-seriated and contains table-shaped cells with an average thickness of 15.63±1.47 µm the cuticle is 14.27±3.4 µm thick, on average; (Figure 2A). *G. brasiliensis* leaves are hypostomatous, showing on average 122±28 stomata mm^{-2} and a ratio between the polar and equatorial stomatal diameters of 1.72±0.14. Stomata are paracytic type (Figure 2C), showing an average length of 37.26±4.07 µm (polar diameter).

The phloem of the midrib is located on both the abaxial and adaxial leaf sides arranged around the xylem in the only vascular bundle, which contains a bundle sheath composed of parenchyma cells (Figure 2B). The midrib and mesophyll regions both contain secretory channels, which show smaller diameters on the adaxial side of the midrib and a large channel on the abaxial side of this region (Figure 2B).

G. brasiliensis primary stems show a one-seriated epidermis with an average thickness of 49.84±10.27 µm; the cuticle has an average thickness of 45.49±07.53 µm (Figures 2D and 2F). Annular collenchyma is found in the external regions right above the epidermis (Figure 2F). Secretory channels are found only in the cortex of stems near the pith (Figures 2D-F). The vascular system at the sampled region of the stem is at the beginning of the development of secondary tissues, showing vascular cambium and secondary xylem and phloem (Figure 2E). The pith parenchyma is composed of round cells showing an average diameter of 63.51±15.71 µm. The cortical parenchyma cells are smaller, showing an average diameter of 30.78±6.38 µm.

G. brasiliensis roots are poliarch, showing seven protoxylem poles interspersed by phloem. Roots show cortex and pith regions that contain round parenchyma cells. A one-seriated endodermis is found around the vascular cylinder. The root pericycle is composed of one to three layers of parenchymatous cells. The root epidermis is one-seriated and the exodermis is composed of one to three layers of parenchymatous cells that contain thicker cell walls compared with the epidermis and cortical parenchyma (Figures 2G and 2H).

4 Discussion

The *G. gardneriana* seeds are exalbuminous and the seed reserves consist of starch grains stored in the ground meristem of the embryos (ASINELLI; SOUZA; MOURÃO, 2011). The same is observed in *G. brasiliensis*, which shows a starch-rich hypocotyl-radicle axis. The starch as the seed reserves may be an apomorphy to the Clusiaceae family and may have taxonomical relevance (CORNER, 1976).

Likewise, the hypocotilar embryo was previously attributed to *Garcinia* plants (NASCIMENTO; CARVALHO; MÜLLER, 2002) and to other Clusiaceae genera such as *Plantonia* (MOURÃO; BELTRATI, 1995) and *Clusia* (MOURÃO; MARZINEK, 2009). According to Mourão and Marzinek (2009), rudimentary cotyledons are also found in *Clusia* embryos and, according to Asinelli, Souza and Mourão (2011), in *Garcinia gardneriana* embryos. Therefore, the hypocotilar embryo, showing rudimentary cotyledons, as found in *G. brasiliensis*, is very similar to many Clusiaceae embryos, confirming an apomorphy of the group.

Clusiaceae seeds may exhibit lipids as the seed storage reserves, as reported for *Vismia guianensis* (Aubl.) Choisy and *Clusia parviflora* Humb. & Bonpl. ex Willd. (MOURÃO; BELTRATI, 2001; MOURÃO; MARZINEK, 2009). Despite this fact, we found that in *G. brasiliensis* the most important seed reserves are comprised of starch grains. The same was reported for *Mammea americana* L. (Clusiaceae) seeds showing starch-rich seeds and lipids detected only in the secretory channel cells (MOURÃO; BELTRATI, 2000). The oleosomes detected in cells of *G. brasiliensis* may be related to defense against herbivores, insects, or pathogens. The well-developed cuticle may be related to the defense against pathogens that may penetrate the inner part of the seed, reaching the embryo.

Phenolics are very common in Clusiaceae species (MOURÃO; BELTRATI, 2000) and were detected and isolated from the *G. brasiliensis* epicarp by chromatography (GONTIJO et al., 2012). However, these compounds were not detected in *G. brasiliensis* embryos via the applied hystochemical test, which may be related to the low

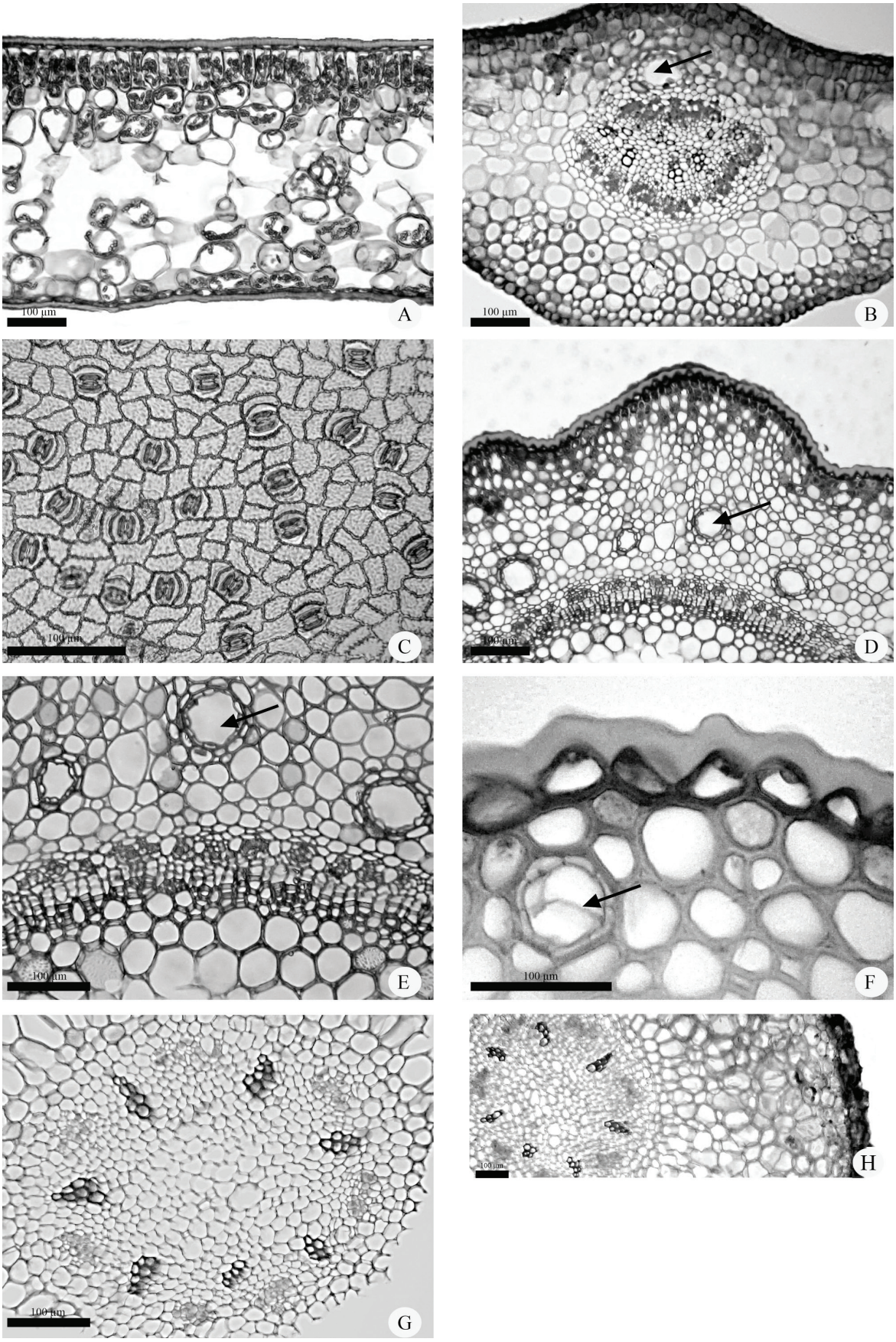

Figure 2. Transversal (A-B; D-F) and paradermal (C) sections showing details of the leaves (A, B, and C), stem (D, E, and F), and roots (G and H) of *Garcinia brasiliensis* plants. Arrows indicate secretory channels. Bars= 100 µm.

concentration of phenolics in plant embryos, not detected by the applied test. Additional tests and detailed extraction may be important to confirm the lack of phenolics in *G. brasiliensis* embryos.

The thickness of the cuticle and epidermis, as well as the sinuosity of the epidermal cell walls, are important traits in Clusiaceae taxonomy (METCALFE; CHALK, 1950). Dorsiventral leaves and a thick cuticle are also reported to Sri Lankan *Garcinia* species (PATHIRANA; HERAT, 2004). There is still little information about Brazilian *Garcinia* species anatomy and physiology. However, Boeger, Alves and Negrelle (2004) reported dorsiventral leaves and a one-seriated epidermis for *G. gardneriana* plants. Likewise, the quantitative anatomical data for leaf tissues reported by these authors are very similar to those found for *G. brasiliensis*.

Quantitative anatomical data may be important to Clusiaceae taxonomy. However, there is little information available concerning the anatomy of this family. The leaf structure and the quantitative anatomical data obtained in this work for *G. brasiliensis* are very similar to those of other *Garcinia* species. Likewise, Pathirana and Herat (2004) reported that quantitative anatomical data may be more important for differentiating groups within *Garcinia*.

Paracytic stomata are very common in the Clusiaceae family (METCALFE; CHALK, 1950). However, this stomatal type may be divided into subtypes: hemyparacytic, showing only one subsidiary cell parallel to the larger stomatal axis (VAN COTTHEM, 1970) and the brachyparcytic type containing two subsidiary cells parallel to the larger stomatal axis (DILCHER, 1974). Pathirana and Herat (2004) reported the hemyparacytic type for *G. morella*, *G. terpnophyla*, *G. thwaitesii*, and *G. zeylanica* and the brachyparacytic type for *G. hermoniii*, *G. quaesita*, *G. terpnophylla*, *G. xanthochymus,* and *G. zeylanica*. Despite the fact that the most common stomatal type in *Garcinia* is paracytic, species of this genus may exhibit anomocytic, tetracytic, sphenocytic, and encyclocytic stomata (PATHIRANA; HERAT, 2004). There is little information about the stomatal type of Brazilian *Garcinia* species and there are no reports for stomatal classification in earlier works. Likewise, *G. brasiliensis* stomata may be classified as brachyparacytic. This may be an important characteristic for further works; therefore, more attention must be devoted to the anatomies of Brazilian species.

The organization of the leaf vascular system in *G. brasiliensis* is very similar to that reported for *Garcinia* species, where the phloem is organized around the xylem in vascular bundles (PATHIRANA; HERAT, 2004). According to Pathirana and Herat (2004), there are six types of organization to the *Garcinia* vascular system in the midrib among Sri Lankan species and this is an important trait for species identification. However, any of these six types of vascular system organizations reported for Sri Lankan species matches the *G. brasiliensis* type. Therefore, *G. brasiliensis,* as a neotropical species, is very different from Asian species and this fact may have evolutionary implications.

The anatomical description of the stem of the *Garcinia* species is still very incipient. In *Calophyllum brasiliense* Cambess (Clusiaceae), the stem structure is very similar to that of *G. brasiliensis*. However, *C. brasiliense* shows a larger number of secretory channels in the cortex (GASPAROTTO-JÚNIOR et al., 2005). The primary root of Clusiaceae plants is poorly developed and becomes degenerate after a few days; as a consequence, adventitious roots are produced, developing the plant root system (NASCIMENTO; CARVALHO; MÜLLER, 2002). There are few works in the literature concerning the root anatomy of Clusiaceae species. Despite the modifications in leaf anatomy of *G. brasiliensis* growing in flooded environments in the Amazon forest (PAROLIN, 2009), the *G. brasiliensis* roots do not show any adaptations related to flooding tolerance. However, modifications in the plant environment may lead to modifications in root anatomy. According to Souza et al. (2013), roots of *G. brasiliensis* seedlings may develop a thicker exodermis, a thicker phloem, a larger number of xylem vessels, and fewer xylem fibers under flooding. Little aerenchyma development was reported by these authors in *G. brasiliensis* roots under flooding. Therefore, *G. brasiliensis* shows root anatomical plasticity and this plasticity may be related to its wide distribution in Brazil.

5 Conclusions

The embryo structure and leaf anatomy of *G. brasiliensis* are very similar to those of other Clusiaceae and *Garcinia* species; however, this species shows differences that may help in plant taxonomy and evolutionary studies. Anatomical works concerning on the Clusiaceae family are very rare and further works may help in the understanding of the taxonomy, phylogeny, physiology, and ecology of this group.

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References

ABDULLAH, N. A. P.; ISMAIL, M. F. Emergence of Multiple Seedlings from Seed of *Garcinia mangostana* L. (Clusiaceae). *Journal of Cell & Plant Sciences*, v. 1, p. 1-5, 2010.

ASINELLI, M. E. C.; SOUZA, M. C.; MOURÃO, K. S. M. Fruit ontogeny of *Garcinia gardneriana* (Planch. & Triana) Zappi (Clusiaceae). *Acta Botanica Brasilica*, v. 25, p. 43-52, 2011.

BOEGER, M. R. T.; ALVES, L. C.; NEGRELLE, R. R. B. Leaf morphology of 89 tree species from a lowland tropical rain forest (Atlantic Forest) in South Brazil. *Brazilian Archives of Biology and Technology*, v. 47, p. 933-943, 2004. http://dx.doi.org/10.1590/S1516-89132004000600013

CAMPANA, R. C.; MOURÃO, K. S. M.; MARZINEK, J. Morfoanatomia e ontogênese dos frutos e sementes de *Clusia lanceolata* Cambess. (Clusiaceae). *Acta Scientiarum - Biological Sciences*, v. 32, p. 437-444, 2010.

CORNER, E. J. H. *The seeds of dicotyledons*. Cambridge: University Press, 1976.

DIAS NETO, O. C.; SCHIAVINI, I.; LOPES, S. F.; VALE, V. S.; GUSSON, A. E.; OLIVEIRA, A. P. Estrutura fitossociológica e grupos ecológicos em fragmento de floresta estacional semidecidual, Uberaba, Minas Gerais, Brasil. *Rodriguésia*, v. 60, p. 1087-1100, 2009.

DILCHER, D. L. Approaches to identification of Angiosperm Leaf remains. *Botanical Review*, v. 40, p. 15-117, 1974. http://dx.doi.org/10.1007/BF02860067

FRANCO, D.; OLIVEIRA, I. V. M.; CAVALCANTE, I. H. L.; CERRI, P. E.; MARTINS, A. B. G. Estaquia como processo de clonagem do bacuripari (*Rheedia gardneriana* Miers ex Planch e Triana). *Revista Brasileira de Fruticultura*, v. 29, p. 176-178, 2007. http://dx.doi.org/10.1590/S0100-29452007000100037

GALETTI, M.; DONATTI, C. I.; PIZO, M. C.; GIACOMINI, H. C. Big Fish are the Best: Seed Dispersal of *Bactris glaucescens* by the Pacu Fish (*Piaractus mesopotamicus*) in the Pantanal, Brazil. *Journal of Tropical Biology and Conservation*, v. 40, p. 386-389, 2008.

GASPAROTTO-JÚNIOR, A.; FERREIRA, I. C. P.; NAKAMURA, C. V.; DIAS FILHO, B. P.; JACOMASSI, E.; YOUNG, M. C. M.; CORTEZ, D. A. G. Estudo morfo-anatômico das folhas e caule da *Calophyllum brasiliense* (Clusiaceae), uma contribuição ao estudo farmacognóstico da droga vegetal. *Acta Farmacológica Boanerense*, v. 24, p. 371-376, 2005.

GONTIJO, V. S.; SOUZA, T. C.; ROSA, I. A.; SOARES, M. G.; SILVA, M. A.; VILEGAS, W.; VIEGAS JÚNIOR, C.; SANTOS, M. H. Isolation and evaluation of the antioxidant activity of phenolic constituents of the *Garcinia brasiliensis* epicarp. *Food Chemistry*, v. 132, p. 1230-1235, 2012. http://dx.doi.org/10.1016/j.foodchem.2011.10.110

HEMSHEKHAR, M.; SUNITHA, K.; SANTHOSH, M. S.; DEVARAJA, S.; KEMPARAJU, S.; VISHWANATH, B. S.; NIRANJANA, S. R; GIRISH, S. K. An overview on genus *Garcinia*: phytochemical and therapeutical aspects. *Phytochemistry Reviews*, v. 10, p. 325-351, 2011. http://dx.doi.org/10.1007/s11101-011-9207-3

KRAUS, J. E.; ARDUIN, M. *Manual básico de métodos em morfologia vegetal*. Seropédica: EDUR, Rio de Janeiro, 1997. 198 p.

LUCAS, C. M. Within flood season variation in fruit consumption and seed dispersal by two characin fishes of the Amazon. *Biotropica*, v. 40, p. 581-589, 2008. http://dx.doi.org/10.1111/j.1744-7429.2008.00415.x

METCALFE, C. R.; CHALK, L. *Anatomy of the dicotyledons*. Claredon Press: Oxford, 1950. 1500 p.

MOURÃO, K. S. M.; BELTRATI, C. M. Morfologia dos frutos, sementes e plântulas de *Platonia insignis* Mart. (Clusiaceae). I. aspectos anatômicos dos frutos e sementes em desenvolvimento. *Acta Amazônica*, v. 25, p. 11-32, 1995.

MOURÃO, K. S. M.; BELTRATI, C. M. Morphology and anatomy of developing fruits and seeds of *Mammea americana* (Clusiaceae). *Revista Brasileira de Biologia*, v. 60, p. 701-711, 2000. http://dx.doi.org/10.1590/S0034-71082000000400023

MOURÃO, K. S. M.; BELTRATI, C. M. Morphology and anatomy of developing fruits and seeds of *Vismia guianensis* (Aubl.) Choisy (Clusiaceae). *Revista Brasileira de Biologia*, v. 61, p. 147-158, 2001. http://dx.doi.org/10.1590/S0034-71082001000100019

MOURÃO, K. S. M.; MARZINEK, J. Fruit ontogenesis in *Clusia parviflora* Humb. & Bonpl. ex Willd. (Clusiaceae). *Acta Botanica Brasilica*, v. 23, p. 797-804, 2009. http://dx.doi.org/10.1590/S0102-33062009000300020

NASCIMENTO, W. M. O.; CARVALHO, J. E. U.; MÜLLER, C. H. Caracterização morfológica da semente e da plântula de bacurizinho (*Rheedia acuminata* (ruiz et pav.) Plachon et triana - Clusiaceae). *Revista Brasileira de Fruticultura*, v. 24, p. 555-558, 2002. http://dx.doi.org/10.1590/S0100-29452002000200055

PAROLIN, P. Submerged in darkness: adaptations to prolonged submergence by woody species of the Amazonian floodplains. *Annals of Botany*, v. 103, p. 359-376, 2009. http://dx.doi.org/10.1093/aob/mcn216

PATHIRANA, P. S. K; HERAT, T. R. Comparative vegetative anatomical study of the genus *Garcinia* L. (Clusiaceae / Guttiferae) in Sri Lanka. *Ceylon Journal of Science*, v. 32, p. 39-66, 2004.

SACRAMENTO, C. K.; COELHO JUNIOR, E.; CARVALHO, J. E. U.; MULLER, C. H.; NASCIMENTO, W. M. O. Cultivo do Mangostão no Brasil. *Revista Brasileira de Fruticultura*, v. 29, p. 195-203, 2007. http://dx.doi.org/10.1590/S0100-29452007000100042

SOUZA, T. C.; SOUZA, E. S.; DOUSSEAU, S.; CASTRO, E. M.; MAGALHÃES, P. C. Seedlings of *Garcinia brasiliensis* (Clusiaceae) subjected to root flooding: Physiological, morphoanatomical, and antioxidant responses to the stress. *Aquatic Botany*, v. 111, p. 43-49, 2013. http://dx.doi.org/10.1016/j.aquabot.2013.08.006

STEVENS, P. F. Clusiaceae-Guttiferae. In: KUBITZKI, K. (Ed.). *The families and genera of vascular plants*. Berlin: Springer-Verlag, 2007.

VAN COTTHEM, W. R. J. A classification of stomatal types. *Botanical Journal of Linnean Society*, v. 63, p. 235-246, 1970. http://dx.doi.org/10.1111/j.1095-8339.1970.tb02321.x

ZAMITH, L. R.; SCARANO, F. R. Produção de mudas de espécies das Restingas do município do Rio de Janeiro, RJ, Brasil. *Acta Botanica Brasilica*, v. 18, 161-176, 2004. http://dx.doi.org/10.1590/S0102-33062004000100014