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Article

Leaf architecture of Rubiaceae Juss. from caatinga vegetation in Brazil

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Abstract: The study and characterization of leaf venation plays a key role in the recognition of taxonomic groups that have been identified mainly based on reproductive traits. This study aimed at characterizing the pattern of leaf venation of Rubiaceae, recognizing individual characters among the taxa of this group and testing the use of leaf architecture as a taxonomic tool capable of providing vegetative and diagnostically characteristics between species. Leaves of 14 species distributed in eight genera from Rubiaceae were diaphanized and classified. The study area is located in Área de Proteção Ambiental Serra Branca/Raso da Catarina, Bahia State, Brazil. The characterization of secondary veins, including type, spacing and number of pairs, third veining patterns, fourth and fifth order, and the conformation of the areola were useful to delimit taxa when used together, providing subsidies to more comprehensive studies.
Keywords: *Diaphanization, Leaf veining, Raso da Catarina, Semiarid.*

Arquitetura foliar de espécies de Rubiaceae Juss. da caatinga brasileira

Resumo: O estudo e a caracterização da venação foliar representam uma possibilidade no reconhecimento de grupos taxonômicos, identificados predominantemente com base em caracteres reprodutivos. O presente estudo objetivou caracterizar o padrão de nervação foliar de Rubiaceae, reconhecer caracteres particulares dentre os táxons deste grupo e testar a utilização da arquitetura foliar como ferramenta taxonômica capaz de fornecer características diagnósticas vegetativas entre as espécies. Foram diafanizadas e classificadas as folhas de 14 espécies distribuídas em 8 gêneros de Rubiaceae na APA Serra Branca/Raso da Catarina. A caracterização das nervuras secundárias, incluindo tipo, espaçamento e número de pares, padrões de nervuras de terceira, quarta e quinta ordem, bem como a conformação das aréolas foram úteis para delimitar os táxons quando utilizados em conjunto, fornecendo subsidío para estudos mais abrangentes.
Palavras-chave: *Diafanização, Nervação foliar, Raso da Catarina, Semiárido.*

Introducion

Rubiaceae Juss. is the fourth largest family in number of species of angiosperms (Robbrecht 1988). In Brazil, the family is represented by 126 genera and 1,412 species, of these 84 genera and 406 species occur in the Northeast, with the greatest diversity in the state of Bahia, where it was recorded 78 genera and 364 species (Barbosa et al. 2018).

Rubiaceae is a monophyletic group of easy circumscription. However, it still faces problems regarding intrafamily classification (Verdcourt 1958, Bremekamp 1966, Bremer & Jansen 1991). The use of vegetative characteristics as diagnostic elements for plant specimens have been considered important in the identification process, in particular those related to the shape of the constituent parts of the plant. Although the shape is accepted as an important element in the taxonomic study, its application has not been explored to its full potential due to the inherent difficulty of description and comparison (Niklas 1994).

Acording to Stace (1989) states that leaf morphology has always played an important role in plant systematics as a whole, particularly to characterize and identify taxa where variation in floral structures is not informative. The leaves present characteristics that can also be used in evolutionary studies, being considered as useful as the floral characteristics, pollen morphology and most of the anatomical characteristics traditionally used in the systematics (Cabral et al. 2016, Ellis et al. 2009, Judd et al. 2009, Hickey & Taylor 1991).

The study of foliar architecture, which was created to assist in the identification of fossils (Hickey 1973), has developed considerably over the years, constituting an important tool for classifying and differentiating problematic groups (Ellis et. al. 2009, Judd et al. 2009, Leaf Architecture 1999).

According to Hickey (1973) the term "leaf architecture" indicates the position and shape of the elements that constitute the external expression of the leave structure including venation pattern, marginal configuration, leaf shape and glandular position, this term is appropriate because the elements of the leaves are arranged in certain defined structural standards that can be described.

Therefore, this study aimed at characterizing the pattern of Rubiaceae leaf venation in the Área de Proteção Ambiental Serra Branca/Raso da Catarina, located in Jeremoabo, Bahia State, recognizing individual characters among the taxa of this group and testing the use of leaf architecture as taxonomic tool capable of providing vegetative diagnostic characteristics among species.

Material and Methods

The Área de Proteção Ambiental Serra Branca/Raso da Catarina (APASB) is located in the municipality of Jeremoabo, northeast of Bahia, between the coordinates 09º53'15.5'' to 09º44'34.6''S and 38º49'36,1'' to 38º52'20,4''W. The area have 67,237 hectares, bordering to the north with the Estação Ecológica Raso da Catarina (ESEC) and to the south with the Vaza-Barris river, which is the main affluent of the São Francisco river in the region. The area is flat with sandstone formations (Szabo et al., 2007).

There were a total of six collection of the botanical material in different days, from May/2014 to June/2015, covering both periods: rainy and hot/dry. The samples obtained in the field were herborized, according to the methodology of Fosberg & Sachet (1965) and Mori et al. (1989), and are deposited in the herbarium of the Universidade do Estado da Bahia (HUNEB - Paulo Afonso Collection). Fully expanded sun leaves of the third and fourth knot were selected for the study. The fresh materials were fixed in 70% FAA according to the methodology described by Johansen (1940), remaining for 72 hours in the solution. Thereafter, the leaves were transferred to 70% ethanol solution (v/v), where they remained preserved until the technical diaphanization procedure.

For each species, 3-4 leaves were selected. The leaves were mostly intact and uninjured. The leaves were diaphanizated in order to not occur any distortion of venation or leaf shape as a whole, resulting from the process of pieces of assembly.

The diaphanization technique used in this work was a modification of the method proposed by Shobe & Lersten (1967). Posteriorly, semipermanent slides with 50% glycerin solution (v/v), and slides with Canada balsam were prepared, and were photographed using a camera (AxioCam ERc5s) attached to the microscope (Zeiss Primo Star). Hickey (1973), Leaf Architecture (1999) and Ellis et al. (2009) were used as references for the description and classification of the veining patterns.

Results

There were a total of 14 species distributed in eight different genera of Rubiaceae of APASB, and described below.

1. *Borreria spinosa* (L.) Cham. & Schltdl.

Symmetrical slide, elliptic to lanceolate, attenuate base, acute apex, entire margin. Pinnate venation, eucamptodromous-brochidodromous. Secondary vein decreasing to the base four-five pairs/side. Intersecondary moderately developed veins, strongly printed. Tertiary veining with alternating percorrent, branched (Figure 1A). Fourth and fifth order reticulated veining in regular polygons. Areola moderately developed, forming three-five sides. Final veins forming one-two branches with no extended vascular endings. Terminal venation of the margin without teeth.

2. *Borreria verticillata* (L.) G. Mey.

Symmetrical and elliptical slide, truncated base, acute apex, serrated margin. Pinnate venation, eucamptodromous. Secondary veins growing to base with three-four pairs/side. Absent intersecondary veins. Randomly reticulated tertiary veins, sinuous, moderately branched. Dichotomized veins of fourth and fifth order (Figure 1B). Undeveloped areolas with three-five sides (Figure 1C). Free final veins forming a branch with enlarged vascular endings (Figure 1D). Terminal venation of the margin forming incomplete ties without teeth.

3. *Cordiera rigida* (K. Schum.) Kuntze

Symmetrical slide elliptic to obovate, acute base, obtuse to subacute apex, entire margin. Pinnate venation, brochidodromous. Secondary veins decreasing to base (Figure 1E) with five-seven pairs/side. Developed intersecondaries veins, heavily printed (Figure 1F). Tertiary opposite percorrent veins, winding. Fourth and fifth order reticulated veins in regular polygons. Developed areolas with four-five sides (Figure 1G). Free final veins forming one-three branches with no extended vascular endings.

4. *Eumachia depauperata* (Müll. Arg.) M.R. Barbosa & M.S. Pereira.

Symmetrical slide, elliptical, convex base, acute apex, entire and revolute margin. Pinnate venation, weak brochidodromous. Irregular secondary veins, with 10 pairs/side. Well developed intersecondaries veins, strongly printed. Tertiary veins randomly reticulated, sinuous, branching. Fourth and fifth order reticulated vein in regular polygons. Areola moderately developed (Figure 1H), with three-five sides. Free final veins forming one-two branches (Figure 1I) with no extended vascular endings. Venation margin of terminal forming ties incomplete without teeth.

5. *Hexasepalum apiculatum* (Willd.) Delprete & J. H. Kirkbr.

Symmetrical slide oblong to lanceolate, acute apex, attenuate base, entire margin. Pinnate venation, eucamptodromous-brochidodromous. Secondary veins decreasing to the base with three pairs/side. Absent intersecondaries veins. Tertiary percorrent alternating veins, winding to ramified. Quaternary reticulated in regular polygons veins. Dichotomized fifth order veins. Undeveloped areolas with four-five sides. Free final veins forming one branch with no extended vascular endings (Figure 1J). Venation margin of terminal forming ties incomplete without teeth.

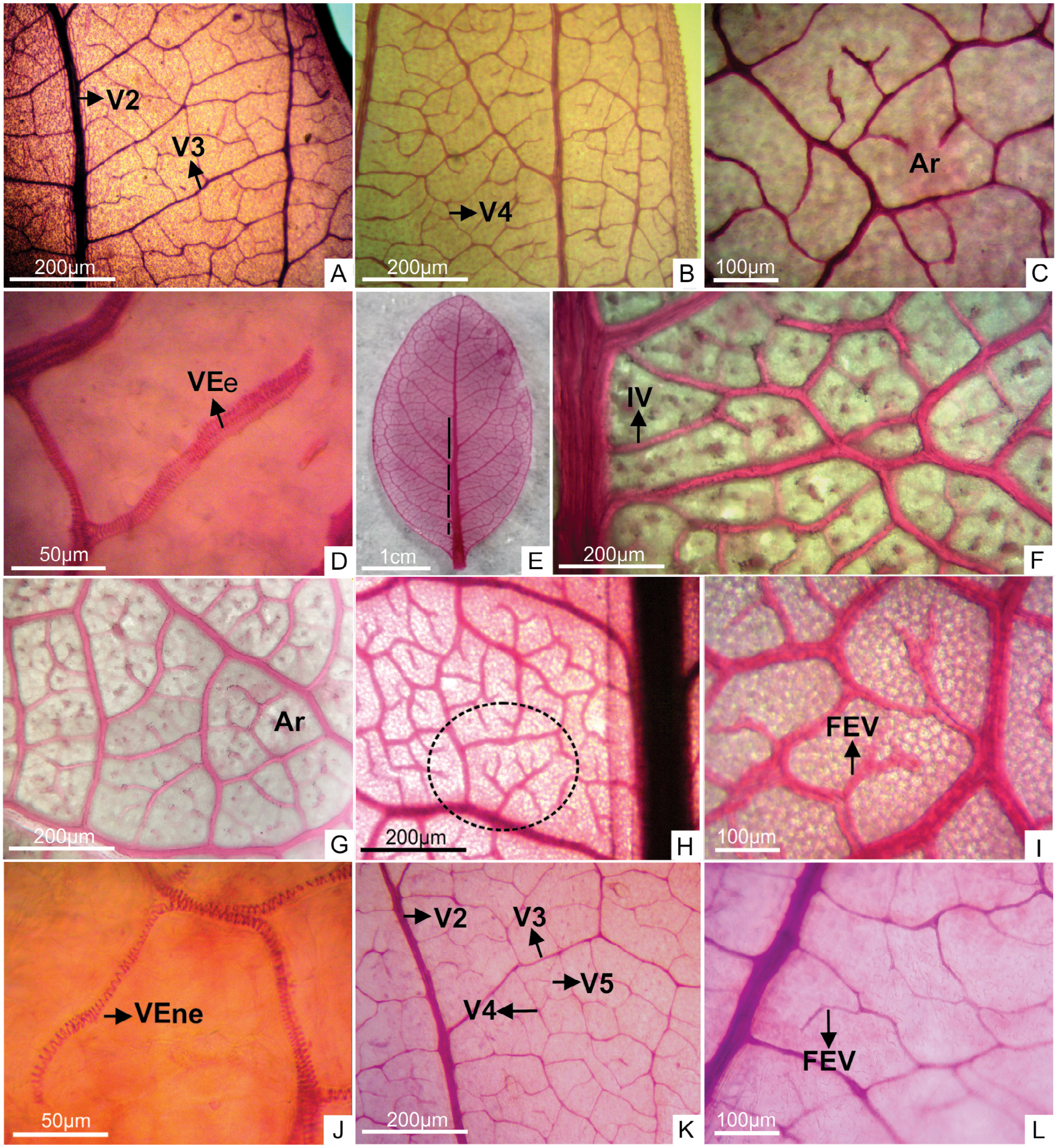

Figure 1. Diaphanized leaves of Rubiaceae of Área de Proteção Ambiental Serra Branca/Raso da Catarina, Bahia. **(A)** *Borreria spinosa*, detail of branched tertiary veins; **(B-D)** *Borreria verticillata*, **B-** with the fourth and fifth order dichotomized veins, **C-** undeveloped areolas and **D-** extended vascular endings; **(E-G)** *Cordiera rigida*, **E-** showing secondary veins decreasing to the base, **F-** intersecondary veins and **G-** developed areolas. **(H)** *Hexasepalum apiculatum*, detail of the final free vein with extensive vascular endings; **(I,J)** *Eumachia depauperata*, **I-** detail of moderately developed areolas and **J-** free end with two branches; **(K,L)** *Mitracarpus salzmannianus*, **K-** detail of the fourth and fifth order reticulated veins in regular polygons and **L-** free end veins with one branch.

6. *Hexasepalum radulum* (Willd.) Delprete & J. H. Kirkbr.

Elliptical slide with asymmetrical base, acute apex, entire margin. Pinnate venation, eucamptodromous. Secondary veins decreasing to base with four-five pairs/side. Absent intersecondaries veins. Mixed opposite/alternating tertiary veins, winding. Quaternary reticulated veins in regular polygons. Areolas developed with three-five sides. Free final veins without branches or forming a (two-three) branch with extended vascular endings. Venation margin of terminal forming ties incomplete without teeth.

7. *Hexasepalum teres* (Walter) J. H. Kirkbr.

Symmetrical slide, lanceolate, attenuate base, acute to acuminate apex, entire margin. Pinnate venation, eucamptodromous-brochidodromous. Growing secondary veins to the base, with 4 pairs/side. Absent intersecondaries veins. Tertiary percorrent opposite veins, winding. Quaternary reticulated veins in regular polygons. Dichotomized fifth order veins. Areolas developed with three-five sides. Free final veins without branches or forming a (one-two) branch with extended vascular endings. Venation margin of terminal forming ties incomplete without teeth.

8. *Mitracarpus baturitensis* Sucre

Symmetrical slide, linear-elliptic, cuneate base, acute apex, entire margin. Pinnate venation, eucamptodromous. Secondary veins decreasing to base with four-five pairs/side. Little evident intersecondaries veins, weak printed. Tertiary opposite percorrent veins, winding. Fourth order reticulated veins in regular polygons. Dichotomized fifth order. Areola moderately developed with four-five sides. Free final veins forming a branch with broad vascular endings. Venation margin of terminal forming ties incomplete without teeth.

9. *Mitracarpus longicalyx* E.B. Souza & M.F. Sales

Symmetrical slide, elliptical, attenuate base, acute-mucronate apex, entire margin. Pinnate venation, eucamptodromous. Secondary veins decreasing to base with five-six pairs/side. Little evident intersecondary veins, weak printed. Opposite/alternating mixed veins, tertiary winding. Fourth and fifth order reticulated veins in regular polygons. Areola moderately developed with four-five sides. Free final one-two veins forming branches with no extended vascular endings. Venation margin of terminal forming ties incomplete without teeth.

10. *Mitracarpus robustus* E.B. Souza & E.L. Cabral

Symmetrical slide, elliptical, attenuate base, acute apex, entire margin. Pinnate venation, eucamptodromous. Secondary veins decreasing to base with five-seven pairs/side. Underdeveloped intersecondary veins, poorly printed. Alternated percorrent tertiary veins, branched. Fourth and fifth order reticulated veins in regular polygons. Undeveloped areolas with three-five sides. Free final one-two veins forming branches with no extended vascular termination. Venation margin of terminal forming ties incomplete without teeth.

11. *Mitracarpus salzmannianus* DC.

Symmetrical slide, elliptical, attenuate base, acute-mucronate apex, entire margin. Pinnate venation, eucamptodromous-brochidodromous. Secondary veins decreasing to the base, with five pairs/side. Intersecondary underdeveloped veins, poorly printed. Percorrent alternate tertiary veins, branched. Fourth and fifth order reticulated veins in regular polygons (Figure 1K). Areola moderately developed with four-five sides. Final ending veins forming an enlarged vascular termination branch (Figure 1L). Venation margin of terminal forming ties incomplete without teeth.

12. *Richardia grandiflora* (Cham. & Schltdl.) Steud.

Symmetrical slide, elliptical, attenuate base, acute apex, entire margin. Pinnate venation, eucamptodromous. Secondary veins growing to the base, with five pairs/side. Absent intersecondary veins. Alternated percorrent tertiary veins, straight, little branched. Dichotomized fourth and fifth order veins. Areola moderately developed with four-five sides. Free final one-two veins forming branches with no extended vascular endings. Venation margin of terminal forming ties incomplete without teeth.

13. *Staelia galioides* DC.

Symmetrical slide, linear-lanceolate, attenuate base, acute apex, entire margin. Pinnate venation, brochidodromous. Secondary veins growing to the base, with four-five pairs/side. Underdeveloped intersecondary veins, poorly printed. Percorrent alternate tertiary veins, branched. Fourth and fifth order reticulated veins in regular polygons. Areola moderately developed, with three-four sides. Free final one-two veins forming branches with no extended vascular endings. Venation margin of terminal forming ties incomplete without teeth.

14. *Tocoyena formosa* (Cham. & Schltdl.) K. Schum

Symmetrical slide, oval to elliptical, cuneate base, acute apex, entire margin. Pinnate venation, brochidodromous. Uniform secondary veins, with nine-twelve pairs/side. Intersecondary, developed veins, heavily printed. Alternated percorrent tertiary veins, straight. Fourth and fifth order reticulated veins in regular polygons. Moderately developed areolas with four-five sides. Free final veins forming one-two branches with no extended vascular endings. Venation margin of terminal forming ties incomplete without teeth.

Key for Rubiaceae species from APA Serra Branca, Bahia, based on standard characters of leaf venation

1. Eucamptodromous venation; terminal venation margin in incomplete ties.
    2. Secondary veining with decreasing spacing to the base.
        3. Vein network forming up to the fourth order; free final veins with up to three branches .... *Hexasepalum radulum*
        3'. Vein network forming up to the fifth order; free final veins with up to two branches.
            4. Mixed opposite/alternating tertiary veins .... *Mitracarpus longicalyx*
            4'. Tertiary opposite percorrent or alternate percorrent veins.
                5. Intersecondary veins absent ...... *Hexasepalum apiculatum*
                5'. Intersecondary veins present.
                    6. Secondary veins with four-five pairs; free final veins with extended vascular endings .... *Mitracarpus baturitensis*
                    6'. Secondary veins with five-seven pairs; free final veins with not extended vascular endings .... *Mitracarpus robustus*

2'. Secondary veins with spacing growing to the base.

7. Secondary veins with three-four pairs; undeveloped areolas; free final veins with extended vascular endings ..... *Borreria verticillata*

7'. Secondary veins with five pairs; moderately developed areola; free final veins with non-extended vascular endings .... *Richardia grandiflora*

1'. Broquidodromous venation, eucamptodromous-brochidodromous or weak brochidodromous; terminal venation of margin in ties.

8. Tertiary randomly reticulated veins .... *Eumachia depauperata*

8'. Tertiary percorrent or alternate percorrent veins.

9. Strong intersecondary veins.

10. Secondary veining with decreasing spacing to the base with five-seven pair; tertiary winding veins .... *Cordiera rigida*

10'. Secondary veins evenly spaced, with nine-twelve pairs; tertiary straight veins .... *Tocoyena formosa*

9'. Weak intersecondary veins, or absent.

11. Undeveloped areolas; free final veins with enlarged vascular endings .... *Hexasepalum teres*

11'. Moderately developed areola; free final veins with no extended vascular endings.

12. Secondary veining with decreasing spacing to the base; free final veins forming a branch ..... *Mitracarpus salzmannianus*

12'. Secondary spaced veins growing to the base; free end veins forming up to two branches.

13. Areolas with up to five sides .... *Borreria spinosa*

13'. Areolas with up to four sides .... *Staelia galioides*

Discussion

The most used vegetative morphological characters for taxa delimitation are the forms, especially the leaves, because it is the growing body of better access. In this study, these elements were not relevant since there was no significant variation between species, characterized primarily by elliptical type, which may be related to genetic issues. This statement agrees with Parkhurst & Loucks (1972) who state that the size and shape of the leaves are controlled by heredity, as demonstrated in their studies, by high variation of types that occur between different species coexisting in a particular environment. Still, it should be noted that the origin of leaf form occurs for a brief morphogenesis initial period, being more influenced by the formation of secondary veins, since the veins of smaller caliber arise only during expansion of the sheet (Dengler & Kang 2001).

The primary standards of leaf architecture of the studied species correspond to the pinned type, where a main vein of larger caliber diverges from the base to the tip of the slide. In general, the primary and secondary veins are the backbones of the leaf, and tertiary veins are on the smaller-caliber veins, which form a kind of mesh or lattice. The primary vein has the highest caliber and generally extends along the leaf, starting from the base or near the base of the leaf and moving toward the margin or apex (Obermüller et al. 2011).

The secondary veins ranged from broquidodromous to eucamptodromous, although this first type is a common standard for Rubiaceae (Mattos 2011, Mantovani et al. 1995, Da Cunha & Vieira 1997), the latter was well represented in this study, constituting 50% of the species studied, Varjão et al. (2013) observed the same parameters. The species *Borreria spinosa*, *Cordiera rigida*, *Hexasepalum teres* e *Mitracarpus salzmannianus* showed a variation between these two types, described as eucamptodromous-brochidodromous, where secondary veins are strongly curved, joining lightly to each other. In addition, *Eumachia depauperata* showed the standard semibroquidodromous, where secondary veins grow towards the margins and unite forming slightly undeveloped arches. The secondary veins are the next set to be measured after the primaries, they often also have a long haul, and that usually goes from the base of the leaf or a primary vein toward the margin or following in arcs towards the apex (Obermüller et al. 2011).

The spacing between the secondary veins varied between species, allowing the delimitation of taxa. *Borreria spinosa* presented secondary veins decreasing to base with four-five pairs/side, whereas in *B. verticillata* they were found growing to the base with three-four pairs/side. The species of the genus *Hexasepalum* showed decreasing spacing to the base with three pairs/side in *H. apiculatum* and four-five pairs/side *H. radulum*. On the other hand, *H. teres* secondary veins were found to grow for four base pairs. All species of the genus *Mitracarpus* presented spacing of secondary veins decreasing to the base, with four-five pairs/side in *M. baturitensis*, five-six pairs/side in *M. longicalyx*, five-seven pairs/side in *M. robustus* and five pairs/side in *M. salzmannianus*.

The other genera, which are represented in this study by one species each, also presented different spacings. In *Cordiera rigida*, secondary veins were falling to the base with five-seven pairs, in *Eumachia depauperata*, the spacing is irregular with ten pairs. *Richardia grandiflora* and *Staelia galioides* showed growing spaces for the base, with five pairs in the representative of the genus *Richardia* and four-five pairs for the kind of *Staelia*. *Tocoyena* presented secondary veins with uniform spacing.

Tertiary veins and smaller orders veins are less visible and keep a similar caliber, forming reticles. Tertiary veins generally have a narrower size than the set of secondary veins and often connect with the primary and secondary veins throughout the sheet (Obermüller et al. 2011). The importance of the tertiary venation pattern is emphasized by Hickey (1973) and Dilcher (1974). The pattern is a dense, well-structured network that supports webs of smaller calibers. The percorrent alternate type was the most representative of the species, which agress to Saha et al. (2014). Tertiary patterns found in this study were essential to distinguish species.

The highest standards of crosslinking are formed until the fifth order of veins for most species herein studied. Only *Hexasepalum radulum* presented veins just until the fourth order. This ratio of the number of cores orders in one species is not linked to the size of the leaf, since relatively large *Mitracarpus* leaf has the same number of cores orders compared to the *Borreria* species, which have tiny slides. These observations are supported by the work of Saha et al. (2014) for Rubiaceae in India. Most species were fourth and fifth reticulated order veins in regular polygons. However, *H. apiculatum*, *H. teres*, *M. robustus* e *M. baturitensis* differed with dichotomized fifth order veins, and *B. verticillata* and *Richardia grandiflora* with both dichotomized orders. It is noteworthy that the higher the density of the veins, more channels per unit area are available for driving, thus helping transpiration and therefore the maintenance of the leaves for water supply (Fonsêca et al. 2006), and sustention (Souza et al. 2013).

Smaller areas formed by tertiary veins and lower orders of veins originate the so-called areolas, which can provide development, arrangement and different form and may or may not present free end veins inside, which may or may not contain branches. They are considered developed when they have mesh size and regular shape; moderately developed, when the meshes vary in size and have an irregular manner; or undeveloped, incomplete or without limiting the sides of the mesh. In this work, the last two types were observed. The areolas are considered diagnosed vegetative characters consistent in a taxon, being very relevant to the taxonomy (Metcalf & Chalk 1950, Solereder 1908). According to Saha et al. (2014), the nature of the final formation of the areola and the endings free veins are of less importance to the family as a whole, including the tribal level classification system. However, in this study, it was observed that the arrangement and shape of the areola were different and unique for each taxon. These characters were important to differentiate taxa, corroborating to the work from Oliveira et al. (2011), where these characters were used to differentiate Myrtaceae species.

The free end veins (FEVs) or free venules are commonly composed by two-three branches. They are thin and can fill the areal space according to their length. The length is not uniform for a species and is randomly oriented (Saha et al. 2014). The species showed mostly FEVs forming one-two branches usually with ramification. However, *Borreria verticillata*, *Hexasepalum apiculatum*, *Eumachia depauperata*, *Mitracarpus baturitensis* e *M. robustus* presented FEVs with only one branch, and *Cordiera rigida* and *H. radulum* with one-three branches.

The free venules have dual function: carry water and dissolved solutes in the transpiration stream and absorb and translocate the products of photosynthesis to other parts of the plant (Menezes et al. 2012). They can also differentiate for presenting or not extended vascular endings and are used in the delimitation of taxa, as stated by Alvarez et al. (2006) and Mattos (2011). In this study the presence of extensive vascular endings were essential to differentiate *Mitracarpus baturitensis* from *M. robustus*, since they present non-extended vascular endings.

The results showed that the standard features of venation when used together serve as a good taxonomic tool for the separation of genera and species within the family Rubiaceae.

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Author Contributions

Mirella Priscila de Souza Lima: Contribuição substancial no conceito e design do estudo, coleta, análise e interpretação de dados, preparação do manuscrito e revisão crítica.

Adriana Soares: Contribuição na coleta, análise e interpretação de dados, e preparação do manuscrito.

José Lucas Ribeiro de Sousa: Contribuição na análise e interpretação de dados.

Márcia Santos Carvalho: Contribuição na coleta, análise e interpretação de dados.

Jorge Marcelo Padovani Porto: Contribuição para revisão crítica, adicionando conteúdo intelectual.

Francyane Tavares Braga: Contribuição no conceito e design do estudo, coleta e análise de dados, e revisão crítica.

Conflicts of interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

References

ALVAREZ, AS, POTIGUARA, RCV, SANTOS, JUM (2006). Arquitetura foliar de espécies de *Eugenia* L. (Myrtaceae), da restinga de Algodoal, Maiandeua, Pará. Bol. Mus. Para. Emilio Goeldi Cienc. Nat., 1 (2): 29-36.

BARBOSA, MR, ZAPPI, D, TAYLOR, C, CABRAL, E, JARDIM, JG, PEREIRA, MS, CALIÓ, MF, PESSOA, MCR, SALAS, R, SOUZA, EB, DI MAIO, FR, MACIAS, L, ANUNCIAÇÃO, EA DA, GERMANO FILHO, P, OLIVEIRA, JA, BRUNIERA, CP, GOMES, M, DE TONI, K, FIRENS, M. 2015. Rubiaceae in Lista de Espécies da Flora do Brasil. Jardim Botânico do Rio de Janeiro. Disponible in: <http://floradobrasil.jbrj.gov.br/jabot/floradobrasil/FB210>. accessed 27 May 2018.

BREMEKAMP, CEB (1966). Remarks on the position, the delimitation and the subdivision of the Rubiaceae. Act. Bot. Nearlandica. 15:1-33.

BREMER, B. & JENSEN, RK (1991). Comparative Restriction Site Mapping of Chloroplast DNA Implies New Phylogenetic Relationships within Rubiaceae. American Journal of Botany. 78 (2):198-213.

CABRAL, LO, FERREIRA, HD, VILHALVA, DAA, FARIA, MT (2016). Arquitetura foliar de espécies arbóreas do bosque Auguste de Saint-Hilaire, Goiânia, Goiás, Brasil. Revista Eletrônica de Educação da Faculdade Araguaia. 9:1-22.

DA CUNHA, M & VIEIRA, RC (1997). Anatomia foliar de *Psychotria velloziana* Benth. (Rubiaceae). Rodriguésia. 45:39-50.

DENGLER, N & KANG, J (2001). Vascular patterning and leaf shape. Plant Biol (Stuttg). 4:50-56.

DILCHER, DL (1974). Approaches to the identification of angiosperm leaf remains. Bot. Rev. 40:1-157.

ELLIS, B, DALY, DC, HICKEY, LJ, JOHNSON, KR, MITCHELL, JD, WILF, P, WING, SL. 2009. Manual of leaf architecture. Ithaca, Cornell University Press.

FONSÊCA, LCM, PROENÇA, CEB, GONÇALVES, EG (2006). Descrição do padrão de venação foliar em *Spathicarpa* Hook. (Araceae). Acta Bot. Bras. 21:213-221.

FOSBERG, FR & SACHET, MH (1965). Manual for tropical herbaria. Netherlands. Utrecht.

HICKEY, LJ 1973. Classification of arquitecture of dicotyledonous leaves. American Journal of Botany. 60 (1):17-33.

HICKEY, LJ & TAYLOR, D. 1991. W.The Leaf architecture of Ticodendron And The Application of foliar Characters In Discerning Its Relationships. Ann. Missouri Bot. Gard. 78:105-130.

JOHANSEN, DA (1940). Plant microtechnique. Mc Graw-Hill. New York.

JUDD, WS, CAMPBELL, CS, KELLOGG, EA, STEVENS, PF, DONOGHUE, MJ. 2009. Sistemática Vegetal: Um Enfoque Filogenético. 3. ed. Porto Alegre: Artmed. 612p

LEAF ARCHITECTURE WORKING GROUP (1999). Manual of leaf Architecture - Morphological description and categorization of dicotyledonous and net-veined monocotyledonous angiosperms. Smithsonian Institution. Washington DC.

MANTOVANI, A, GOMES, M, GOMES, DMS e VIEIRA, RC (1995). Anatomia foliar de *Rudgea decipiens* Müll. Arg. and *R. macrophylla* Benth. (Rubiaceae). Acta Bot. Bras. 9:247-261.

MATTOS, KLBL (2011). Anatomia foliar aplicada à taxonomia em Rubiaceae Juss. Tese. Universidade Federal de Viçosa.

MENEZES, NL, SILVA, DC e MELO DE PINNA, GFA. (2012). Folha. In: APEZZATO-DA-GLÓRIA, B. & CARMELO-GUERREIRO, SM. (Orgs.) Anatomia Vegetal. 3ªed. Ed. UFV. Universidade Federal de Viçosa. Viçosa.

METCALF, CR & CHALK, L. (1950). Anatomy of the dicotyledons: leaves, stem, and wood in relation to taxonomy with notes on economic uses. Oxford. Clarendon.

MORI, SA, SILVA, LAM, LISBOA, GE e CORADIN L. (1989). Manual de Manejo do Herbário Fanerogâmico. Centro de Pesquisa do Cacau. Ilhéus.

NIKLAS, KJ (1994). Plant allometry: the scaling ofform and processo The University of Chicago. Chicago.

OBERMÜLLER, FA, DALY, DC, OLIVEIRA, EC, SOUZA, HFTP; OLIVEIRA, HM, SOUZA, LS e SILVEIRA, M. (2011). Guia Ilustrado e Manual de Arquitetura Foliar para Espécies Madeireiras da Amazônia Ocidental. GK Noronha, Rio Branco, 101p.

OLIVEIRA, MIU, FUNCH, LS, SANTOS, FAR e LANDRUM, LR. 2011. Aplicação de caracteres morfoanatômicos foliares na taxonomia de *Campomanesia* Ruiz & Pavón (Myrtaceae). Acta Bot. Bras. 25: 455-465.

PARKHURST, DF & LOUCKS, OL (1972). Optimal leaf size in relation to environment. J. Ecol. 60:505-537.

ROBBRECHT, E (1988). Tropical woody Rubiaceae Characteristic features and progressions. Contributions to a new subfamilial classification. Opera Bot. Belg. 1:1-271.

SAHA D, MAITI GG, MUKHERJEE SK (2014). Leaf venation pattern of some Indian members of Rubiaceae. IJPRS, 3 (4): 160-169.

SHOBE, WR & LERSTEN, NR (1967). A technique for clearing and staining Gymnosperm leaves. Bot. Gaz. 128:150-152.

SOLEREDER, H. (1908). Systematic anatomy of the dicotyledons. Oxford. Clarendon Press. 1182p.

SOUZA, VC, FLORES, T B, LORENZI, H (2013). Introdução à botânica: morfologia. Nova Odessa- SP, Instituto Plantarum, 224p.

STACE, CA. 1989. Plant Taxonomy and Biosystematics. 2 edição. Cambridge. University Press.

SZABO, AV, ROCHA, ACS, TOSATO, JAC e BARROSO W (2007). Área de Proteção Ambiental (APA) Serra Branca-Raso da Catarina. In: Marques J. (Org.) As Caatingas: debates sobre a Ecorregião do Raso da Catarina. 1ªed. Fonte Viva, Paulo Afonso, pp. 21-39.

VARJÃO, RR, JARDIM, JG e CONCEIÇÃO, AS (2013). Rubiaceae Juss. de caatinga na APA Serra Branca/Raso da Catarina, Bahia, Brasil. Biota Neotrop. 13 (2):105-123.

VERDECOURT, B (1958). Remark on the classification of the Rubiaceae. Bull. Jard. De Etat. Bruxellles 28 (3): 209-281.

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