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ACANTHACEAE

Dicliptera mucronifolia Nees, 2*n* = 60; Brazil, Paraíba, *J.M.P. Cordeiro 320.*

Hygrophila paraibana Rizzini, 2*n* = 60; Brazil, Paraíba, *J.M.P. Cordeiro 1033.*

Justicia chamaedryoides (Nees) Wassh. ex A.L.A.Côrtes & P.L.R. Moraes, 2*n* = 18; Brazil, Paraíba, *J.M.P. Cordeiro 1035.*

Ruellia bahiensis (Nees) Morong, 2*n* = 34; Brazil, Paraíba, *J.M.P. Cordeiro 1021.*

Ruellia inundata Kunth, 2*n* = 34; Brazil, Paraíba, *J.M.P. Cordeiro 1022.*

CACTACEAE

Cereus jamacaru DC., 2*n* = 22; Brazil, Pernambuco, *E.M. Almeida 1082.*

CAPPARACEAE

Neocalyptrocalyx longifolium (Mart.) Cornejo & Iltis, 2*n* = 16; Brazil, Paraíba, *J.M.P. Cordeiro 1023.*

EUPHORBIACEAE

Euphorbia comosa Vell., 2*n* = 40; Brazil, Paraíba, *J.M.P. Cordeiro 1034.*

FABACEAE

Calliandra surinamensis Benth., 2*n* = 16; Brazil, Paraíba, *J.M.P. Cordeiro 1024.*

Inga laurina (Sw.) Willd., 2*n* = 26; Brazil, Paraíba, *L.P. Felix 12260.*
2*n* = 26+1B; Brazil, Paraíba, *L.P. Felix 14860.*
2n = 52; Brazil, Rio Grande do Norte, *L.P. Felix 14605.*

Pithecellobium dulce (Roxb.) Benth., 2*n* = 26; Brazil, Paraíba, *J.M.P. Cordeiro 1038.*

MALVACEAE

Wissadula amplissima (L.) R.E.Fr., 2*n* = 14; Brazil, Paraíba, *J.M.P. Cordeiro 1026.*

ORCHIDACEAE

Habenaria josephensis Barb.Rodr., 2*n* = 50; Brazil, Paraíba, *E.M. Almeida 801.*

Oeceoclades maculata (Lindl.) Lindl., 2*n* = 52; Brazil, Piauí, *E.M. Almeida 1098.*

Phragmipedium sargentianum (Rolfe) Rolfe, 2*n* = 22; Brazil, Bahia, *E.M. Almeida 1006.*

Vanilla pompona Schiede, 2*n* = 32; Brazil, Paraíba, *E.M. Almeida 825.*

PASSIFLORACEAE

Turnera subulata Sm., 2*n* = 40; Brazil, Bahia, *J.M.P. Cordeiro 198.*

SMILACACEAE

Smilax brasiliensis Spreng., 2*n* = 32; Brazil, Rio Grande do Norte, *L.P. Felix 14574.*

VITACEAE

Cissus decidua Lombardi, 2*n* = 34; Brazil, Pernambuco, *J.M.P. Cordeiro 401.*

All materials for the chromosome column should be submitted electronically to: Karol Marhold, karol.marhold@savba.sk (Institute of Botany, Slovak Academy of Sciences, SK-845 23 Bratislava, Slovakia, and Department of Botany, Charles University, CZ 128-01 Prague, Czech Republic). The full version of this contribution is available in the online edition of TAXON appended to this article. The following citation format is recommended: Baltisberger, M. & Voelger, M. 2006. *Sternbergia sicula*. In: Marhold, K. (ed.), IAPT/IOPB chromosome data 1. *Taxon* 55: 444, E2.

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LORANTHACEAE
Tribe Psittacantheae
Subtribe Ligarinae

Ligaria cuneifolia (Ruiz & Pav.) Tiegh., $n = 10$; Brazil, Rio Grande do Sul, *G.A. Dettke & L.F. Lima 171* (ICN), *G.A. Dettke & L.F. Lima 176* (ICN), *G.A. Dettke & L.F. Lima 177* (ICN).

Ligaria teretiflora (Rizzini) Kuijt, $n = 10$; Brazil, Bahia, *M.J.G. Andrade 252* (HUEFS), *M.J.G. Andrade 262* (HUEFS), *M.J.G. Andrade 279* (HUEFS).

Subtribe Psittacanthinae

Psittacanthus bicalyculatus (Mart.) Mart., $n = 8$; Brazil, Bahia, *M.J.G. Andrade 268* (HUEFS), *M.J.G. Andrade 633* (HUEFS).

Struthanthus martianus Dettke & Waechter, $n = 8$; Brazil, São Paulo, *A.P. Moraes 114* (HCF); *A.P. Moraes 120* (BOTU). $2n = 16$; Brazil, São Paulo, *A.P. Moraes 131* (BOTU).

Struthanthus syringifolius (Mart.) Mart., $n = 8$; Brazil, Paraíba, *L.P. Félix 9625* (EAN).

Tripodanthus acutifolius (Ruiz & Pav.) Tiegh., $n = 8$; Brazil, Bahia, *M.J.G. Andrade 267* (HUEFS), *M.J.G. Andrade 634* (HUEFS), *M.J.G. Andrade 280* (HUEFS), *M.J.G. Andrade 368* (HUEFS); Brazil, Rio Grande do Sul, *G.A. Dettke & A.P. Moraes 175* (ICN). $n = 16$; Brazil, Abaíra, *M.J.G. Andrade 368 & al.* (HUEFS).

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BRASSICACEAE

Alyssum murale subsp. *pichleri* (Vel.) Stoj. & Stef., $2n = 48$; Bulgaria, *DP-16023.*

Erysimum scoparium (Wild.) Wettst., $2n = 28$; Canary islands, Tenerife, *DP-16024.*

FABACEAE

Astragalus hamosus L., $2n = 46$; Greece, *DP-15020.*

Astragalus monspessulanus subsp. *illyricus* (Bernh.) Chater, $2n = 16$; Bulgaria, *DP-16001.* $2n = 16+1B$; Bulgaria, *DP-15016.*

PLANTAGINACEAE

Plantago lanceolata L., $2n = 12$; Bulgaria, *DP-16021*, *DP-16022.*

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All materials CHN; collectors: *C* = M.A.N. Coelho, *F* = L.P. Felix, *G* = M. Guerra, *M* = S.J. Mayo, *R* = F.C. Ramalho.

ARACEAE

Aglaonema commutatum Schott, $2n = 120$; Brazil, Pernambuco, *R 766-A* (PEUFR).

Alocasia macrorrhizos (L.) G.Don, $2n = 28$; Brazil, Pernambuco, *R 42* (PEUFR).

Anthurium affine Schott, $2n = 30$; Brazil, Pernambuco, *R 765* (PEUFR); Brazil, Pernambuco, *R 832* (PEUFR).

Anthurium bromelicola Mayo & L.P.Felix, $2n = 30$; Brazil, Pernambuco, *M & F 1156* (EAN).

Anthurium gracile (Rudge) Schott, $2n = 40$; Brazil, Pernambuco, *R & G 744* (PEUFR); Brazil, Pernambuco, *F 12964* (EAN).

Anthurium jilekii Schott, $2n = 30$; Brazil, Pernambuco, *N & al. 874* (PEUFR).

Anthurium pentaphyllum (Aubl.) G.Don, $2n = 30$; Brazil, Paraíba, *F 13663* (EAN); $2n = 60$; Brazil, Pernambuco, *M & al. 907* (PEUFR).

Anthurium petrophilum K.Krause, $2n = 30$; Brazil, Pernambuco, *R 13* (PEUFR); $2n = 30$; Brazil, Paraíba, *F 6174* (EAN).

Anthurium scandens (Aubl.) Engl., $2n = 48$; Brazil, Pernambuco, *R 22* (PEUFR).

Asterostigma riedelianum (Schott) Kuntze, $2n = 34$; Brazil, Pernambuco, *M 1044* (UFP).

Colocasia esculenta (L.) Schott, $2n = 28$; Brazil, Pernambuco, *M 1043* (UFP).

Dieffenbachia seguine (Jacq.) Schott, $2n = 34$; Brazil, Pernambuco, *R 04* (PEUFR).

Dracontioides desciscens (Schott) Engl., 2*n* = 26; Brazil, Sergipe, *F 12933* (EAN).
Dracontium nivosum (Lem.) G.H.Zhu, 2*n* = 26; Brazil, Pará, *F 12682* (EAN).
Monstera adansonii subsp. *klotzschiana* (Schott) Mayo & I.M. Andrade, 2*n* = 60; Brazil, Paraíba, *F 13679* (EAN).
Montrichardia linifera (Arruda) Schott, 2*n* = 48; Brazil, Pernambuco, *R & G 20* (PEUFR).
Philodendron acutatum Schott, 2*n* = 32; Brazil, Pernambuco, *R 30* (PEUFR).
Philodendron bipinnatifidum Schott ex Endl., 2*n* = 36; Brazil, Pernambuco, *R 26* (PEUFR).
Philodendron blanchetianum Schott, 2*n* = 34; Brazil, Pernambuco, *R 27* (PEUFR).
Philodendron fragrantissimum (Hook.) G. Don, 2*n* = 46; Brazil, Pernambuco, *R 30* (PEUFR).
Philodendron hederaceum (Jacq.) Schott, 2*n* = 32; Brazil, Pernambuco, *R 785* (PEUFR).
Philodendron leal-costae Mayo & G.M.Barroso, 2*n* = 36; Brazil, Pernambuco, *R 11* (PEUFR).
Philodendron ornatum Schott, 2*n* = 34; Brazil, Pernambuco, *R 02* (PEUFR).
Philodendron pedatum (Hook.) Kunth, 2*n* = 32; Brazil, Pernambuco, *R 834* (PEUFR).
Philodendron rudgeanum Schott, 2*n* = 40; Brazil, Pernambuco, *R 01* (PEUFR).
Philodendron ruthianum Nadruz, 2*n* = 32; Brazil, Pernambuco, *R 25* (PEUFR).
Pistia stratiotes L., 2*n* = 28; Brazil, Paraíba, *F 10766* (EAN).
Spathicarpa hastifolia Hook., 2*n* = 34; Brazil, Paraíba, cultivated *F 14854* (EAN).
Syngonium podophyllum Schott, 2*n* = 26; Brazil, Pernambuco, *N & al. 884* (PEUFR).
Typhonium roxburghii Schott, 2*n* = ca. 68; Brazil, Pernambuco, *G 856* (PEUFR).
Xanthosoma sagittifolium (L.) Schott, 2*n* = 29; Brazil, Pernambuco, *G 1121* (UFP).
Zomicarpa pythonium (Mart.) Schott, 2*n* = 20; Brazil, Pernambuco, *G 927* (UFP).

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SOLANACEAE

Capsicum parvifolium Sendtn, 2*n* = 24; Brasil, Paraíba, *F.A. Agra & G.E. Barboza 7075* (JPB).

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Joel M.P. Cordeiro, Sarah Nascimento, Angeline M.S. Santos, Enoque Medeiros Neto, Amanda Santos, Felipe Nollet M. de Assis,* Leonardo P. Felix

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Methods for chromosome analysis are according to Guerra & Souza (2002).

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* First chromosome count for the genus.
** First chromosome count for the species.
*** New cytotype for the species.

ACANTHACEAE

***Dicliptera mucronifolia* Nees
2*n* = 60, CHN. Brazil, Paraíba, Maturéia, 07°15′29″ S; 37°23′10″ W, 29 Jul 2014, *J.M.P. Cordeiro 320* (EAN) [Fig. 1A, 2A].

***Hygrophila paraibana* Rizzini
2*n* = 60, CHN. Brazil, Paraíba, Serra da Raiz, 06°43′57″ S, 35°27′25″ W, 6 Mar 2016, *J.M.P. Cordeiro 1039* (EAN) [Fig. 1B, 2B].

***Justicia chamaedryoides* (Nees) Wassh. ex A.L.A.Côrtes & P.L.R.Moraes
2*n* = 18, CHN. Brazil, Paraíba, Pilões, 06°42′00″ S; 35°36′54″ W, 14 Feb 2016, *J.M.P. Cordeiro 1041* (EAN) [Fig. 1C, 2C].

***Ruellia bahiensis* (Nees) Morong
2*n* = 34, CHN. Brazil, Paraíba, Serra da Raiz, 06°43′57″ S, 35°27′25″ W, 21 Nov 2015, *J.M.P. Cordeiro 1021* (EAN) [Fig. 1D, 2D].

Ruellia inundata Kunth
2*n* = 34, CHN. Brazil, Paraíba, Serra da Raiz, 06°43′57″ S, 35°27′25″ W, 21 Nov 2015, *J.M.P. Cordeiro 1022* (EAN) [Fig. 2E].

CACTACEAE

Cereus jamacaru DC.
2*n* = 22, CHN. Brazil, Paraíba, Pernambuco, São Lourenço da Mata, 08°00′13″ S, 35°01′17″ W, 16 Mar 2014, *E.M. Almeida 1082* (EAN) [Fig. 2F].

CAPPARACEAE

**Neocalyptrocalyx longifolium* (Mart.) Cornejo & Iltis
2*n* = 16, CHN. Brazil, Paraíba, Serra da Raiz, 06°43′57″ S, 35°27′25″ W, 21 Nov 2015, *J.M.P. Cordeiro 1023* (EAN) [Fig. 1E, 2G].

EUPHORBIACEAE

Euphorbia comosa Vell.
2*n* = 40, CHN. Brazil, Paraíba, Serra da Raiz, 06°40′44″ S, 35°26′23″ W, 21 Mar 2016, *J.M.P. Cordeiro 1034* (EAN) [Fig. 2H].

FABACEAE

Calliandra surinamensis Benth.
2*n* = 16, CHN. Brazil, Paraíba, Serra da Raiz, 06°40′44″ S, 35°26′23″ W, 21 Nov 2015, *J.M.P. Cordeiro 1024* (EAN) [Fig. 2I].

Inga laurina (Sw.) Willd.
2*n* = 26, CHN. Brazil, Paraíba, Itapororoca, Jacoca, 06°48′51″ S, 35°17′54″ W, 5 May 2008, *L.P. Felix 12260* (EAN) [Fig. 2J].
2*n* = 26+1B, CHN. Brazil, Paraíba, Itapororoca, Jacoca, 06°48′51″ S, 35°17′54″ W, 26 Mar 2014, *L.P. Felix 14860* (EAN) [Fig. 2K].
2*n* = 52, CHN. Brazil, Rio Grande do Norte, Martins, 06°05′12″ S, 37°54′32″ W, 17 Dec 2013, *L.P. Felix 14605* (EAN) [Fig. 3A].

Pithecellobium dulce (Roxb.) Benth.
2*n* = 26, CHN. Brazil, Paraíba, Areia, 06°57′48″ S, 35°41′30″ W, 11 Apr 2016, *J.M.P. Cordeiro 1038* (EAN) [Fig. 3B].

MALVACEAE

Wissadula amplissima (L.) R.E.Fr.
2*n* = 14, CHN. Brazil, Paraíba, Serra da Raiz, 06°43′57″ S, 35°27′25″ W, 21 Nov 2015, *J.M.P. Cordeiro 1026* (EAN) [Fig. 3C].

ORCHIDACEAE

Habenaria josephensis Barb.Rodr.
2*n* = 50, CHN. Brazil, Paraíba, Areia, 06°57′48″ S, 35°41′30″ W, 16 Aug 2013, *E.M. Almeida 801* (EAN) [Fig. 3D].

Oeceoclades maculata (Lindl.) Lindl.
2*n* = 52, CHN. Brazil, Piauí, Pedro II, 04°25′23″ S, 41°27′34″ W, 19 Apr 2014, *E.M. Almeida 1098* (EAN) [Fig. 3E].

Phragmipedium sargentianum (Rolfe) Rolfe
2*n* = 22, CHN. Brazil, Bahia, Santa Terezinha, 12°51′04″ S, 39°28′51″ W, 22 Jan 2014, *E.M. Almeida 1006* (EAN) [Fig. 3F].

Vanilla pompona Schiede
2*n* = 32, CHN. Brazil, Paraíba, Barra de Santana, 07°29′01″ S, 36°02′59″ W, 22 Jul 2013, *E.M. Almeida 825* (EAN) [Fig. 3G].

PASSIFLORACEAE

*** *Turnera subulata* Sm.

2*n* = 40, CHN. Brazil, Bahia, Ibicoara, 12°43′32″ S, 41°17′46″ W, 27 Jan 2014, *J.M.P. Cordeiro 198* (EAN) [Fig. 1G, 3I].

SMILACACEAE

** *Smilax brasiliensis* Spreng.

2*n* = 32, CHN. Brazil, Rio Grande do Norte, Natal, 05°51′28″ S, 35°11′43″ W, 29 Nov 2013, *L.P. Felix 14574* (EAN) [Fig. 1F, 3H].

VITACEAE

Cissus decidua Lombardi

2*n* = 34, CHN. Brazil, Pernambuco, Buíque, 08°35′37″ S, 37°12′20″ W, 30 Jul 2014, *J.M.P. Cordeiro 401* (EAN) [Fig. 3J].

The use of the fluorochromes Chromomycin A3 (CMA) and 4′6-diamidino-2-phenylindole (DAPI) allows to stain differentially genome regions rich in both GC and AT base pairs, respectively (Guerra, 2000; Barros e Silva & Guerra, 2010). The pattern of heterochromatic bands resulting from the use of these fluorochromes enables to differentiate plant groups with less variable chromosome numbers and morphology (Carvalho & al., 2005; Oliveira & al., 2015). Thus, approaches involving fluorochrome staining are generally directed to the analyses of closely related species, as we can see in several works for the genera *Spondias* L. (Almeida & al., 2007), *Epidendrum* L. (Pessoa & al., 2014), and *Zephyranthes* Herb. (Felix & al., 2011), which have the same chromosome numbers, but present variability in heterochromatin band patterns. On the other hand, analysis of CMA/DAPI band patterns in broader taxonomic categories may reveal characteristic patterns in groups of hierarchical levels above genus (Guerra, 2000; Oliveira & al., 2015). This study aimed to document the chromosome number variation and characterize the CMA/DAPI band patterns in species belonging to different plant families, in order to identify heterochromatin band patterns characteristic for some of these groups.

Root tips were pretreated with 8-hydroxyquinoline 0.002 M at 4°C for 24 h, fixed in Carnoy 3 : 1 absolute ethanol/glacial acetic acid (v/v) for 3 h at room temperature and subsequently stored in freezer at −20°C. To prepare the slides, the root tips were washed twice in distilled water and digested in an enzymatic solution containing 2% cellulase and 20% pectinase, and kept in a moist chamber at 37°C for 1 h. The material was squashed in 45% acetic acid, frozen in liquid

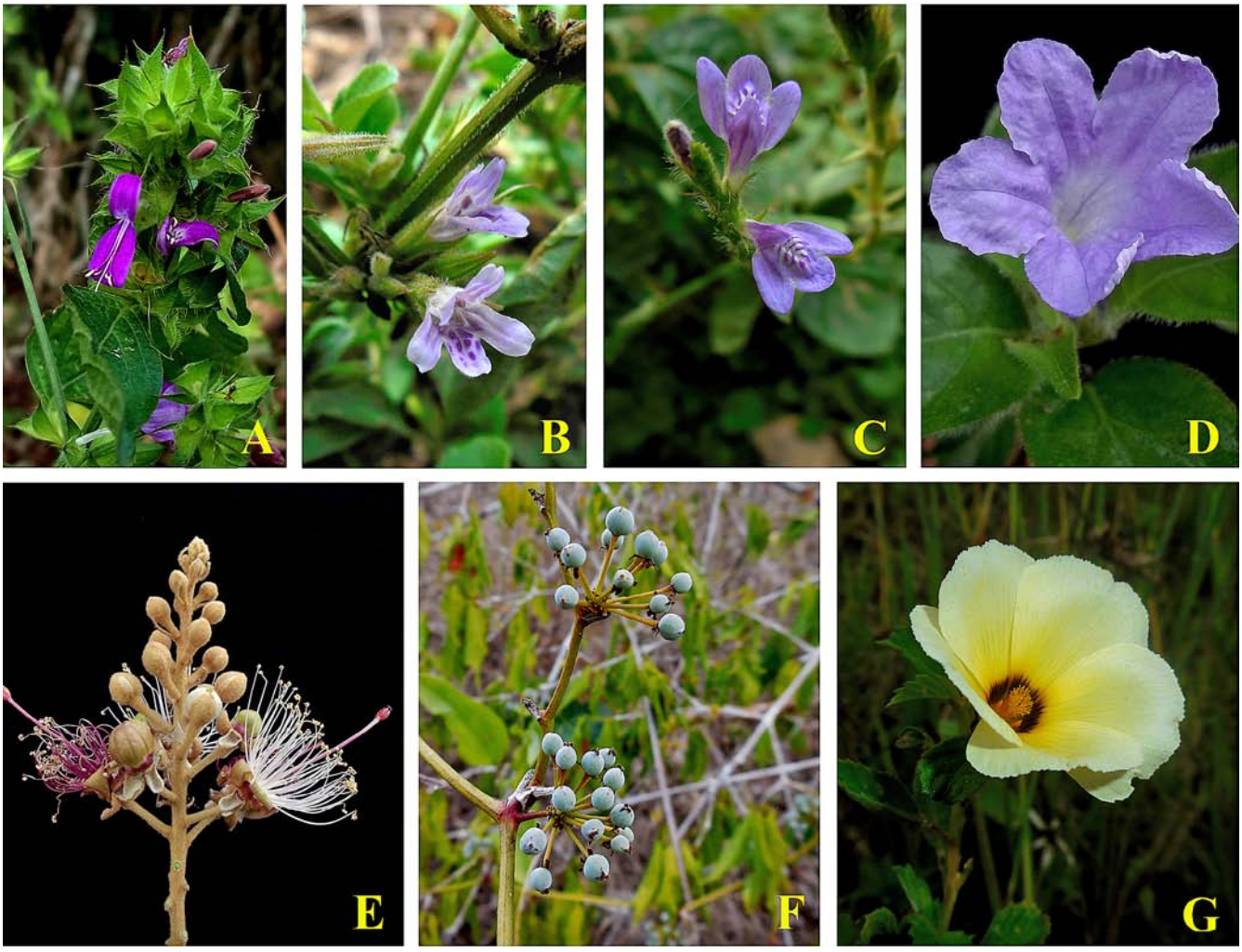

Fig. 1. Species with new chromosome records or cytotypes described. **A,** *Dicliptera mucronifolia*; **B,** *Hygrophila paraibana*; **C,** *Justicia chamaedryoides*; **D,** *Ruellia bahiensis*; **E,** *Neocalyptrocalyx longifolium*; **F,** *Smilax brasiliensis*; **G,** *Turnera subulata*.

nitrogen for remove the coverslip. The slides were then stained with DAPI solution (2 mg/ml) : glycerol (1 : 1, v/v) in order to select the best slides. They were subsequently destained in ethanol-acetic acid (3 : 1) for 30 min at room temperature and then kept in absolute ethanol at 4°C for 2 h. After being dried, the slides were aged for three days at room temperature and then stained for 1 h with 10 µl CMA (0.1 mg/ml) and then with 10 µl DAPI (2 mg/ml) for 30 min, mounted in glycerol/McIlvaine buffer (pH 7.0) (1 : 1, v/v) and then stored for three days in the dark for fluorochromes stabilization (Guerra & Souza, 2002). The best metaphases were captured in photomicroscope Zeiss with Axio Cam MRC5 using Axiovision v.4.8 software. The images were edited using Adobe Photoshop CS3 Extended Version 10.0 software.

New chromosome counts were recorded for the genus *Neocalyptrocalyx* Hutch. (*N. longifolium* (Mart.) Cornejo & Iltis, $2n = 16$), and for the species *Dicliptera mucronifolia* Nees, *Hygrophila paraibana* Rizzini, both with $2n = 60$, *Justicia chamaedryoides* (Nees) Wassh. ex A.L.A.Côrtes & P.L.R.Moraes ($2n = 18$), *Ruellia bahiensis* (Nees) Morong ($2n = 34$) and *Smilax brasiliensis* Spreng. ($2n = 32$) (Fig. 1A–F). The other species had their previous counts confirmed, except *Turnera subulata* Sm. (Fig. 1G), whose chromosome number $2n = 40$ diverged from previous counts with $2n = 10$, 20 (Lopez & al., 2011). In the family Fabaceae Lindl., *Inga laurina* (Sw.) Willd. presented three different cytotypes: an individual with $2n = 26$ (Jacoca population, Fig. 2J), other individual at the same population with $2n = 26+1B$ and other individual with $2n = 52$ (Martins population, Fig. 3A).

For the family Acanthaceae Juss., *D. mucronifolia* possessed two large CMA+ proximal bands and two small CMA+ terminal bands (Fig. 2A), *H. paraibana* possessed six CMA+ terminal bands (Fig. 2B) and *Ruellia inundata* Kunth four CMA+ bands, two large terminal bands and two small proximal bands (Fig. 2E). Two CMA+ bands corresponding to terminal NORs were observed in *J. chamaedryoides* (Fig. 2C) and *R. bahiensis* (Fig. 2D). A pair of CMA+ terminal bands was observed in *Cereus jamacaru* DC. (Fig. 2F) of the family Cactaceae Juss., in *Pithecellobium dulce* (Roxb.) Benth. (Fig. 3B) of the family Fabaceae, in *Wissadula amplissima* (L.) R.E.Fr. (Fig. 3C) of the family Malvaceae Juss., *S. brasiliensis* (Fig. 3H) of the family Smilacaceae Vent., and in *Cissus decidua* Lombardi (Fig. 3J) of the family Vitaceae Juss. On the other hand, *N. longifolium* (Capparaceae Juss.) was the only species that possessed a pair of CMA+ proximal bands extensively distended (Fig. 2G). For the species *I. laurina*, the cytotype with $2n = 26$ possessed two heteromorphic CMA+ terminal bands, while the cytotype with $2n = 26+1B$, besides the two bands corresponding to the NORs, possessed a completely CMA+/DAPI− heterochromatic B chromosome (Fig. 2K, arrow), and small CMA+ proximal bands visualized on prometaphase. The cytotype with $2n = 52$ possessed six CMA bands, with two small terminal bands (Fig. 3A) and four large terminal bands. Largely distended CMA+ terminal bands were observed in *Calliandra surinamensis* Benth. that also possessed four small CMA terminal bands (Fig. 2I). For the family Orchidaceae Juss. were observed CMA+ bands forming large terminal blocks on all (or almost all) chromosomes of *Habenaria josephensis* Barb.Rodr. (Fig. 3D), *Phragmipedium sargentianum* (Rolfe) Rolfe (Fig. 3F) and *Vanilla pompona* Schiede (Fig. 3G), while *Oeceoclades*

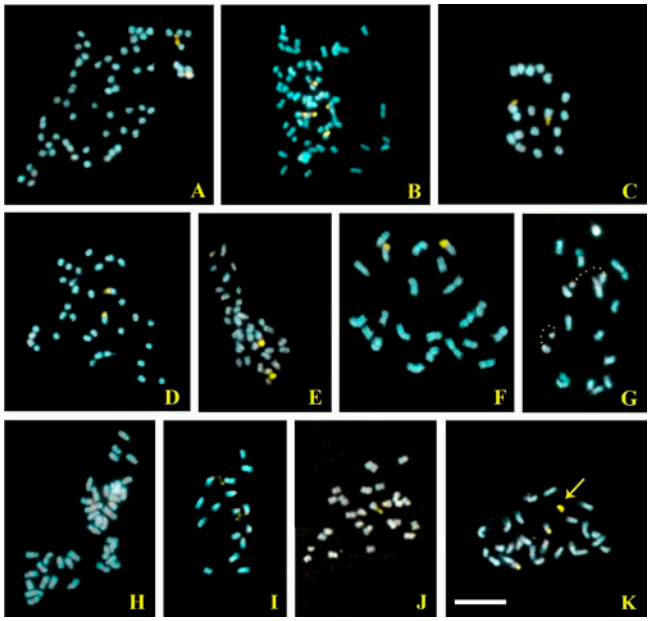

Fig. 2. A, *Dicliptera mucronifolia*, $2n = 60$; **B,** *Hygrophila paraibana*, $2n = 60$; **C,** *Justicia chamaedryoides*, $2n = 18$; **D,** *Ruellia bahiensis*, $2n = 34$; **E,** *Ruellia inundata*, $2n = 34$; **F,** *Cereus jamacaru*, $2n = 22$; **G,** *Neocalyptrocalyx longifolium*, $2n = 16$; **H,** *Euphorbia comosa*, $2n = 40$; **I,** *Calliandra surinamensis*, $2n = 16$; **J,** *Inga laurina*, $2n = 26$; **K,** *I. laurina*, $2n = 26+1B$, arrow indicates B chromosome. — Scale bar = 10 µm.

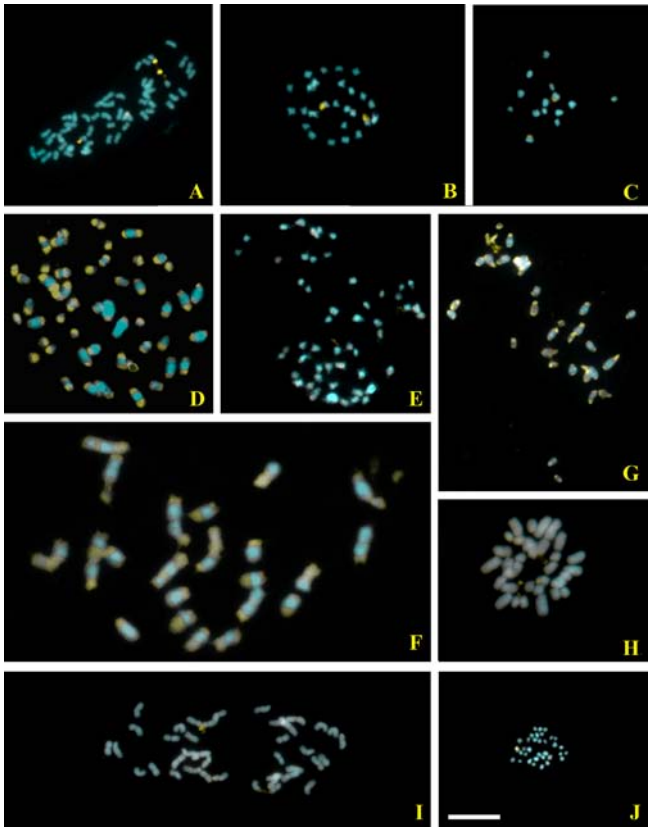

Fig. 3. A, *Inga laurina*, $2n = 52$, CMA band largely distended; **B,** *Pithecellobium dulce*, $2n = 26$; **C,** *Wissadula amplissima*, $2n = 14$; **D,** *Habenaria josephensis*, $2n = 50$; **E,** *Oeceoclades maculata*, $2n = 52$; **F,** *Phragmipedium sargentianum*, $2n = 22$; **G,** *Vanilla pompona*, $2n = 32$; **H,** *Smilax brasiliensis*, $2n = 32$; **I,** *Turnera subulata*, $2n = 40$; **J,** *Cissus decidua*, $2n = 34$. — Scale bar = 10 µm.

maculata (Lindl.) Lindl. possessed only small CMA+ terminal bands on three chromosome pairs, and a CMA+ proximal band on other pair (Fig. 3E). *Euphorbia comosa* Vell. (Euphorbiaceae Juss.), besides two CMA+ terminal bands, also possessed DAPI+/CMA− terminal blocks on both arms from 38 of the 40 chromosomes of this species (Fig. 2H).

For all 19 species analyzed here, no previous records for heterochromatin band patterns were available. The most common pattern, a single CMA+ terminal band by monoploid complement, was observed in 12 of the 19 species studied. This is the more frequent pattern of CMA/DAPI bands in angiosperms and generally corresponds to the GC-rich heterochromatic NORs (reviewed by Guerra, 2000). The occurrence of a large number of CMA bands is generally the result of satellite DNA amplification mediated by unequal crossing-over with gene conversion (Eickbush & Eickbush, 2007), DNA monomers amplification and homogenization by extrachromosomal circular DNA molecules (eccDNA, "rolling circle") in recombination (Cohen & al., 2010), or mediated by retrotransposons (Hobza & al., 2015). It is likely that the amplification of GC-rich subterminal heterochromatin observed here for representatives of the subfamilies Vanilloideae Szlach. (*V. pompona*), Cypripedioideae Garay (*P. sargentianum*) and Orchidoideae Lindl. (*H. josephensis*), as well as the large AT-rich terminal blocks of *E. comososa*, is the result of the associated or isolated action of these mechanisms (Emadzade & al., 2014).

For the subfamily Cypripedioideae, a large number of rDNA sites presumably associated to GC-rich heterochromatin was previously observed for the genus *Paphiopedilum* Pfitzer (Lan & Albert, 2011), whose origin seems related to mechanisms of double-stranded DNA breakage and repair (double-strand break repair) probably mediated by unequal crossing-over. However, the mechanisms involved in the preferential location of large GC-rich (Orchidaceae analyzed here) or AT-rich (*E. comosa*) heterochromatic blocks on the chromosome terminals were not have been identified yet.

As expected, the CMA/DAPI band patterns observed for the species from 10 different angiosperm families (2 monocotyledonous and 8 eudicotyledons) were quite variable, but with a predominance of GC-rich bands on the chromosome terminals of most species, with some species also presenting a small number of subtelomeric or pericentomeric additional bands.

The pattern characterized by large heterochromatic terminal blocks in most chromosomes, observed here in three different subfamilies of orchids, was previously registered in some representatives of the subtribe *Maxillariinae* Benth. (Cabral & al., 2006), as in *Phalaenopsis* Blume (Kao & al., 2001), in *Cypripedium* L. (Kondo & al., 1994), and *Paphiopedilum* Pfitzer (Karasawa & Tanaka, 1980). The widest distribution of this band pattern seems to characterize different genera from Cypripedioideae, and it can be a synapomorphy for the subfamily. On the other hand, for the subfamily Vanilloideae and the genus *Euphorbia* L., previous records for other species involving fluorochrome staining are not known. In Orchidoideae this heterochromatic pattern seems to be restricted to some groups of *Habenaria* Willd. (Felix, 2001), and has not been detected in other genera from this subfamily (D'Emerico & al., 2005). From the chromosome dataset obtained by CMA/DAPI banding in a sample of different angiosperm families, it can be concluded that representatives of the subfamilies Cypripediioideae and Vanilloideae (at least *Vanilla* Mill.) are groups potentially characterized by the presence of large heterochromatic terminal blocks on most of their chromosomes. This pattern, although present in one species of the subfamily Orchidoideae and in the genus *Euphorbia*, needs to be evaluated in a larger sample in order to assess the importance of this character for the cytotaxonomy in these groups.

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* First chromosome count for the species.

LORANTHACEAE
Tribe Psittacantheae
Subtribe Ligarinae

Ligaria cuneifolia (Ruiz & Pav.) Tiegh.

n = 10, CHN. Brazil, Rio Grande do Sul, Bagé, Passo do Batalha, approx. 15 km of the city, 31°23′ S, 54°11′ W, 20 Mar 2009, *G.A. Dettke & L.F. Lima 171* (ICN) [Fig. 4A]; Brazil, Rio Grande do Sul, Bagé, Estância Cerro Alegre, 31°14′24″ S, 53°59′39″ W, 22 Mar 2009, *G.A. Dettke & L.F. Lima 176* (ICN); Brazil, Rio Grande do Sul, Bagé, Igrejinha, approx. 10 km of the city, 31°23′ S, 54°11′ W, 22 Mar 2009, *G.A. Dettke & L.F. Lima 177* (ICN) [Fig. 4B].

**Ligaria teretiflora* (Rizzini) Kujit

n = 10, CHN. Brazil, Bahia, Morro do Chapéu, Chapada Diamantina, between cities Morro do Chapéu and Irecê, at Estrada do Feijão (BA-052), 11°31′09″ S, 41°17′10″ W, 1152 m, 1 May 2003, *M.J.G. Andrade 252* (HUEFS) [Fig. 4C]; Brazil, Bahia, Chapada Diamantina, Morro do Chapéu, Lajedo Bordado, at Formosa locality, 11°16′20.7″ S, 41°05′05.5″ W, 2 May 2003, *M.J.G. Andrade 262* (HUEFS) [Fig. 4D]; Brazil, Bahia, Chapa Diamantina, Morro do Chapéu, at Dunas, 21 km west of the city, 11°29′45″ S, 41°19′54″ W, 9 May 2003, *M.J.G. Andrade 279* (HUEFS) [Fig. 4E].

Subtribe Psittacanthinae

Psittacanthus bicalyculatus (Mart.) Mart.

n = 8, CHN. Brazil, Bahia, Morro do Chapéu, Lajedo Bordado, at Formosa locality, Chapada Diamantina, 11°16′17.8″ S, 41°04′31.3″ W, 773 m, 2 May 2003, *M.J.G. Andrade 268 & al.* (HUEFS) and 5 May 2007, *M.J.G. Andrade 633 & al.* (HUEFS) [Fig. 4F].

Struthanthus martianus Dettke & Waechter

n = 8, CHN. Brazil, São Paulo, Botucatu, Cachoeira da Pavuna, road SP-300, hemiparasitic on *Persea america* Mill. in front of the waterfall trail, 22°50′26.85″ S, 48°30′45.32″ W, 759 m, 19 Dec 2013, *A.P. Moraes 114* (HCF); Brazil, São Paulo, Botucatu, São Paulo State University/UNESP, Campus Rubião Júnior, Botany Depto., 22°53′13.76″ S, 48°29′49.84″ W, 890 m, 10 Jan 2014, *A.P.Moraes 120* (BOTU) [Fig. 4K].

2*n* = 16, CHN. Brazil, São Paulo, Campinas, District of Barão Geraldo, Coco Square at Street Manoel Antunes Novo, 22°49′18.05″ S, 47°04′57.68″ W, 600 m, 21 Jun 2010, *A.P.Moraes 131* (BOTU) [Fig. 4L].

Struthanthus syringifolius (Mart.) Mart.

n = 8, CHN. Brazil, Paraíba, João Pessoa, at Cemitério Senhor da Boa Sentença, 07°07′26.96″ S, 34°53′30.87″ W, 21 m, *L.P. Félix 9625* (EAN).

Tripodanthus acutifolius (Ruiz & Pav.) Tiegh.

n = 8, CHN. Brazil, Bahia, Morro do Chapéu, Lajedo Bordado, at Formosa locality, Chapada Diamantina, 11°16′20.7″ S, 41°05′05.5″ W, 751 m, 2 May 2003, *M.J.G. Andrade 267 & al.* and 5 May 2007, *M.J.G. Andrade 634 & al.* (HUEFS) [Fig. 4H]; Brazil, Bahia, Morro do Chapéu, Lajes locality, at the Dunas, Chapada Diamantina, 11°29′49.7″ S, 41°19′49.6″ W, 944 m, 9 May 2003, *M.J.G. Andrade 280 & al.* (HUEFS) [Fig. 4I]; Brazil, Rio Grande do Sul, Porto Alegre, hemiparasitic on *Ligustrum japonicus* Thunb. (Oleraceae) at Street Veador Porto, close to São Luíz Street, 30°03′00.14″ S, 51°11′58.57″ W, 9 m, 23 Mar 2009, *G.A. Dettke & A.P. Moraes 175* (ICN) [Fig. 4J].

**n* = 16, CHN. Brazil, Abaíra, Catolés, at Boa Vista, Chapada Diamantina, 13°17′54.0″ S, 41°50′42.7″ W, 1069 m, *M.J.G. Andrade 368 & al.* (HUEFS) [Fig. 4G].

Loranthaceae Juss. is the largest family of Santalales with 77 genera encompassing 950 species distributed mainly throughout the tropical and subtropical regions (Nickrent & al., 2010; Kujik & Hansen, 2015). Chromosome numbers are conserved in monophyletic clades and $x = 12$ is suggested as the basic chromosome number (Vidal-Russel & Nickrent, 2008; Kujik & Hansen, 2015). Aneuploidy is the main mechanism of chromosome evolution, with four major events in the family (Nickrent & al., 2010). The tribes Nuytsieae Tiegh., Gaiadendreae Tiegh. and Elytrantheae Engl., all sister of Psittacantheae Horan., and the subtribes Notantherinae Nickrent & Vidal-Russell and Tupeinae Nickrent & Vidal-Russell (both in the tribe Psittacantheae) present $x = 12$. The two first descending aneuploidies are observed inside the tribe Psittacantheae: the first in the subtribe Ligarinae Nickrent & Vidal-Russell, with $x = 10$; and the second, in the subtribe Psittacanthinae Engl., with $x = 8$. The third and the fourth descending

aneuploidies occurred in the tribe Lorantheae Rchb.: the third event occurred in the subtribe Ileostylinae Nickrent & Vidal-Russell ($x = 11$), and the fourth in the clade comprising subtribes Loranthinae Engl., Amyeminae Nickrent & Vidal-Russell, Scurrulinae Nickrent & Vidal-Russell, Dendrophthoinae Nickrent & Vidal-Russell, Emelianthinae Nickrent & Vidal-Russell and Tapinanthinae Nickrent & Vidal-Russell ($x = 9$).

Despite the large size of the chromosomes, just a low proportion of the Brazilian Loranthaceae has been analysed. According to Arruda & al. (2012), 131 species of Loranthaceae occur in Brazil, but only 19 of them have their chromosome number published (see Andrade & al., 2005). Probably, the absence of roots, which are modified in haustorium, prevents the karyotype analysis and the chromosome data available are usually based on meiotic analysis. The mitotic analysis in this family depends on embryos, but the mucilage present in the fruit hamper the chromosome spread. Due to these difficulties, until now no chromosome banding or in situ hybridization data are available to this family.

Here we confirm the chromosome counts for five species, based on thirteen populations distributed from southern to northern Brazil. We also present a new ploidy level for *T. acutifolius* (tetraploid) and the chromosome count for *L. teretiflora*. The meiotic normality (five species) and pollen viability (four species) were estimated and chromosome banding, for *L. cuneifolia,* and in situ hybridization, for *T. acutifolius* and *S. martianus*, are presented. The meiotic and pollen grain preparations were performed following Moraes & al. (2015)

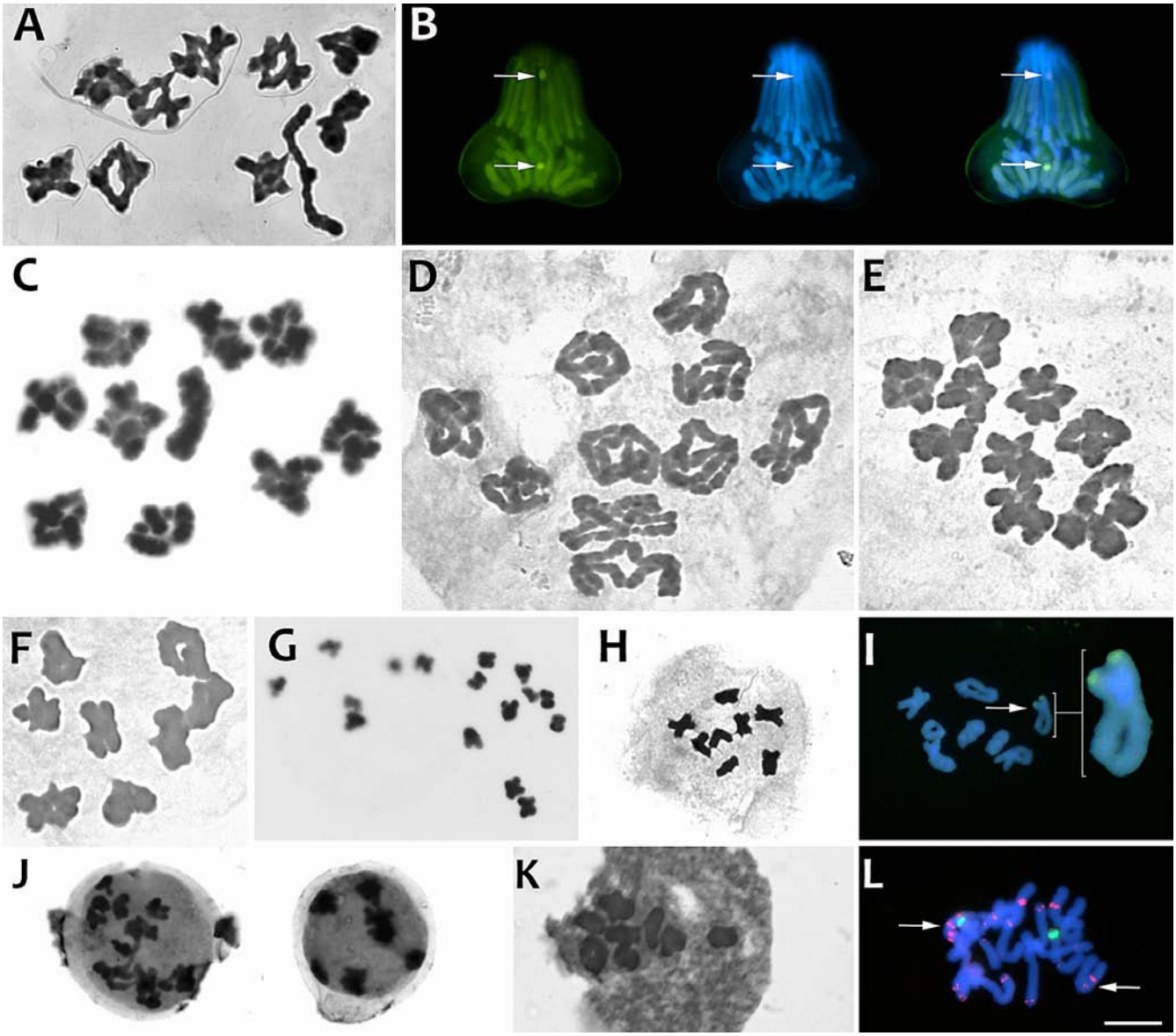

Fig. 4. Loranthaceae chromosomes. **A,** *Ligaria cuneifolia*, $n = 10$; **B,** *L. cuneifolia*, anaphase in the first pollen grain mitosis; **C–E,** *L. teretiflora*, $n = 10$; **F,** *Psittacanthus bicalyculatus*, $n = 8$; **G,** Tetraploid *Tripodanthus acutifolius*, $n = 16$; **H & I,** Diploid *T. acutifolius*, $n = 8$; **J,** Meiotic error in *T. acutifolius*; **K,** *Struthanthus martianus*, $n = 8$; **L,** *S. martianus*, $2n = 16$. — Arrows in B and I indicate the CMA^+ bands. Insert in I shows the homologous pair with terminal CMA^+ enlarged. The metaphase in L shows the localization of 45S rDNA sites (green) and 5S rDNA sites (red, arrows). — Scale bar = 10 µm.

with minor modifications. For meiosis preparations, the anthers were hydrolysed in HCl 1N at 60°C for 5 min and squashed in a drop of warm 60% acetic acid. If material were available, additional slides were prepared for chromosome banding. The anthers were washed two times in distilled water and digested in a solution of 2% (w/v) cellulase (Onozuka)/20% (v/v) pectinase (Sigma)/1% macerozyme (Sigma) at 37°C for 5 min. The anthers were squashed in a drop of warm 60% acetic acid and after removing the coverslip in liquid nitrogen, the slides were aged for three days and stained following Schweizer (1976). Mitotic preparation of *S. martianus* used embryos. The pre-treatments and fixation followed Guerra & Souza (2002), with minor modifications. The fixative solution was composed by 9 : 3 : 1 (absolute alcohol : glacial acetic acid : chloroform, v : v : v). In situ hybridization followed Schwarzacher & Heslop-Harrison (2000).

All slides were analysed under a microscope BX51 (Olympus) coupled with CCD digital camera Evolution MT using Image ProPlus v.6 software (Media Cybernetics) and the captured images were edited for equally brightness and contrast using Adobe Photoshop CS5. The meiotic normality and pollen viability were compared using a parametric analysis of variance test followed by a Tukey test using BioEstat v.5.0 (Ayres & al., 2007).

The chromosome number observed in *L. cuneifolia* and *L. teretiflora* was $n = 10$ and in *P. bicalyculatus*, *T. acutifolius*, *S. martianus* and *S. syringifolius* it was $n = 8$. One individual from *T. acutifolius* represented a tetraploid cytotype with $n = 16$.

All species analysed here are included in two subtribes of the tribe Psittacantheae:

Subtribe Psittacanthinae (11 genera with 247 spp., $x = 8$): chromosome numbers have been reported for 36 species from eight genera (Covas, 1949; Hunziker & Perez-Moreau, 1961; Wiens, 1964; Kuijt, 1975; Barlow & Wiens, 1971; Andrade & al., 2005), all presenting $n = 8$. Polyploidy was detected just twice in this subtribe: in the tetraploid *Passovia pyrifolia* (Kunth) Tiegh. from Costa Rica (Barlow & Wiens, 1971) and in the tetraploid individual of *T. acutifolius* from Catolés/Bahia (present work).

Subtribe Ligarinae (2 genera with 13 spp., $x = 10, 12$): chromosome number of $n = 10$ have been reported for both species of *Ligaria* – *L. cuneifolia* (Barlow & Wiens, 1971; Covas & Schnack, 1946) and *L. teretiflora* (present work). The remaining 11 species are grouped in the genus *Tristerix* Mart., with four species analysed, all presenting $n = 12$.

The meiotic analysis showed a high normality among the 13 populations from the five analysed species (Table 1). However, differences were found among *T. acutifolius* populations ($F = 7.9199$, $p = 0.0075$): all populations from Northeast were localized in the countryside and presented a higher normality compared with the population from South, collected in the city (*Dettke & Moraes 175*). The bioassay studies using the epiphytic *Tradescantia* L. in the same city showed a negative effect of pollution in the microsporogenesis, increasing the number of micronuclei (Costa & Droste, 2012). The pollution could be the cause of the meiotic abnormality observed in *Tripodanthus*. However, even in the case of increase of meiotic errors, the average normality was always above 90%. Andrade & al. (2005) also found a low percentage of meiotic abnormalities (10%) among the ten analysed species, mainly precocious segregation and anaphase bridges with fragment – the same abnormalities found here (Fig. 4J). The pollen viability (estimated by stainability with carmim) was equally high in all populations ($F = 0.7378$, $p = 0.6603$), even considering just *T. acutifolius* populations ($F = 0.7978$, $p = 0.5545$).

Considering the chromosome banding, a similar result was found in both species analysed – one CMA^+ band per haploid genome (arrows in Fig. 4B and I). The CMA^+ band is a CG-rich heterochromatic block, as the 45S rDNA site, and, usually, both sites are co-localized (Roa & Guerra, 2012). For *S. martianus*, one 45S rDNA pair of site was found in the diploid genome (green blocks in Fig. 4L), suggesting that the pattern of CMA^+ and 45S rDNA site distribution could be similar among the three species, *S. martianus, L. cuneifolia* and *T. acutifolius* (Fig. 4B, I & L). The presence of one terminal 45S rDNA pair, as observed in *S. martianus*, is the commonest condition in angiosperms (Roa & Guerra, 2012). The 45S rDNA sites tend to vary more in number than the 5S rDNA sites (Roa & Guerra, 2012, 2015), in contrast to observed here in *S. martianus*. Fourteen 5S rDNA sites were distributed across 12 chromosomes (red blocks in Fig. 4L), one chromosome pair presenting duplicated sites (see arrows in Fig. 4L). If on one hand, *S. martianus* did not show less 5S rDNA sites than 45S rDNA sites; on the other hand, the position of the sites agrees with the hypothesis of Roa & Guerra (2015), who suggested that, in large chromosomes (>6 µm), 5S rDNA sites are located in terminal/interstitial positions. The same pattern of number and position of 5S rDNA sites was found for *P. bicalyculatus* (A.P. Moraes, data not shown).

Our results reaffirm the stability of the chromosome number in Loranthaceae, what is correlated with the subtribes organization. The preliminary results of chromosome banding and in situ hybridization also suggest stable karyotypes in the family, even the peculiar pattern of 5S rDNA could be found in the two distinct genera. Besides, the relationship of higher microsporogenesis error and air pollution suggests that mistletoe could be useful to environmental bio-monitoring, as commonly done with *Tradescantia* L. (Mišík & al., 2007, 2011). However, such plants have an advantage: large and beautiful chromosomes, making the challenging microsporogenesis analysis much easier.

Table 1. Meiosis normality and pollen viability in Loranthaceae.

Species	Meiosis normality (%)*	Pollen viability (%)
Ligaria cuneifolia (Ruiz & Pav.) Tiegh.		
G.A. Dettke & L.F. Lima 171 (ICN)	100	98.07
Psittacanthus bicalyculatus (Mart.) Mart.		
M.J.G. Andrade 268 (HUEFS)	99.36	98.93
M.J.G. Andrade 633 (HUEFS)	94.93	98.50
Struthanthus martianus Dettke & Waechter		
A.P. Moraes 114 (HCF)	97.00	–
A.P. Moraes 120 (BOTU)	99.33	–
Struthanthus syringifolius (Mart.) Mart.		
L.P. Félix 9625 (EAN)	95.67	99.27
Tripodanthus acutifolius (Ruiz & Pav.) Tiegh.		
M.J.G. Andrade 267 (HUEFS)	99.80[a]	98.87
M.J.G. Andrade 280 (HUEFS)	97.17[a]	98.93
M.J.G. Andrade 368 (HUEFS)	98.60[a]	–
M.J.G. Andrade 634 (HUEFS)	98.50[a]	98.58
G.A. Dettke & A.P. Moraes 175 (ICN)	95.73[b]	99.00

* For *T. acutifolius*, means followed by the same letter are not significantly different ($p < 0.05$) based on Tukey test.

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All plant material originates from natural habitats. All cytological studies have been carried out on root tips of seedlings, pretreated in 0.2% colchicine, fixed in ethanol-acetic acid (3 : 1) and stained in hematoxylin (Pearse, 1962). Nomenclature for chromosome morphology follows Levan & al. (1964).

* First chromosome count for the subspecies.
▼ First chromosome count from serpentine area.

BRASSICACEAE

Alyssum murale subsp. *pichleri* (Vel.) Stoj. & Stef.

▼2*n* = 6*x* = 48, CHN. Bulgaria, Central Rhodope Mts., southward from Parventez village, on serpentine rocks, 278 m, 42°03′57.74″ N, 24°39′26.24″ E, 15 Aug 2016, *D. Pavlova DP-I6023* (SO) [Fig. 5A].

Alyssum murale is a polymorphic species widely distributed in Europe, including Bulgaria. Two subspecies (subsp. *murale* and subsp. *pichleri*) are recognized for the flora of Bulgaria (Stojanov, 1970; Anchev, 2007) both distributed on and off serpentines. These taxa are also well known as Ni hyperaccumulators (Bani & al., 2010). The chromosome number previously reported for *A. murale* from different localities of its area (Goldblatt & Johnson 1979–; Anchev, 1991; 2001; Warwick & Al-Shehbaz, 2006) is 2*n* = 16. According to Anchev (1991; 2001) *A. murale* subsp. *pichleri* is diploid (2*n* = 16). The hexaploid chromosome number 2*n* = 48 is reported here for the first time for populations of *A. murale*.

Erysimum scoparium (Wild.) Wettst.

2*n* = 28, CHN. Canary Islands, Tenerife, Canadas del Teide, 2138 m, 28°13′23.94″ N, 16°37′51.11″ W, 18 Oct 2016, *D. Pavlova DP-I6024* (SO) [Fig. 5B].

The chromosome number is congruent with the reports summarized by Rice & al. (2015) for this endemic plant.

FABACEAE

Astragalus monspessulanus subsp. *illyricus* (Bernh.) Chater

2*n* = 16, CHN. Bulgaria, Znepole floristic region, northwest of the town of Tran, the gorge of the Erma River, calcareous rocks, 683 m,

42°51′37.32″ N, 22°38′46.27″ E, with fruits, 10 Jul 2016, *D. Pavlova DP-16001* (SO) [Fig. 5C].

* 2*n* = 16+1B, CHN. Bulgaria, Western Stara planina Mts., in the region of chalet Parshevitza, calcareous rocks on Krastanova mogila peak, 1379 m, 43°08′04.22″ N, 23°29′04.25″ E, 13 Jul 2015, *D. Pavlova DP-15016* (SO) [Fig. 5D].

This subspecies is very rare in the Bulgarian flora and is only known from two localities at a distance of about 100 km from each other, at different altitudes and floristic regions. The karyotype is symmetrical and consists of metacentric and submetacentric chromosomes (Fig. 5C). The diploid chromosome number reported by Druskovic & Lovka (1995) for this subspecies is confirmed. The chromosome number 2*n* = 16+1B is reported for the first time for this subspecies (Fig. 5D).

Astragalus hamosus L.

2*n* = 46, CHN. Greece, northwestward from Ammoudia village, near to the sea shore, 11 m, 39°14′30.14″ N, 20°28′27.52″ E, 4 Sep 2015, *D. Pavlova DP-15020* (SO) [Fig. 5E]

The previous karyological studies showed a varying chromosome number for this species: 2*n* = 24, 32, 40, 44, 46, 48 (Horjales, 1976); 2*n* = 40, 42, 44, 46, 48 (Pavlova, 1995); 2*n* = 44 (Luque & Lifante, 1991). This count is different from the previously reported 2*n* = ca. 44 (Runemark, 2006) for the populations of the species from Greece (Kiklades, Naxos, Oros Zeus). The examined species population was aneuploid. The chromosomes are relatively small (ca. 1 µm) and their morphology is not clear. Dane & al. (2007) considered mitotic irregularities the main reason for aneuploidy and different chromosome numbers reported.

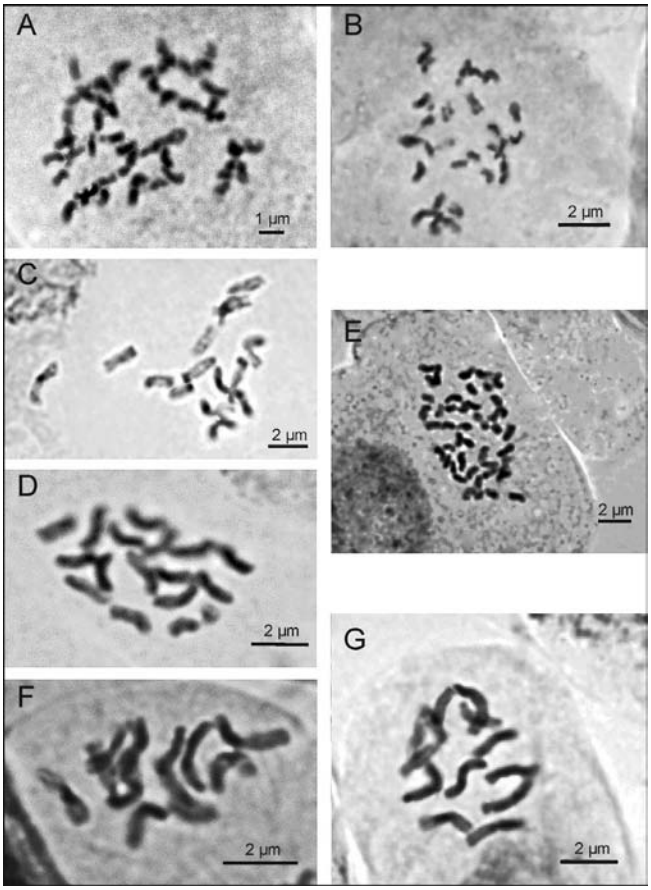

Fig. 5. Metaphase chromosome plate of: **A,** *Alyssum murale* subsp. *pichleri* (*DP-16023*); **B,** *Erysimum scoparium* (*DP-16024*); **C,** *Astragalus monspessulanus* subsp. *illyricus* (*DP-16001*); **D,** *Astragalus monspessulanus* subsp. *illyricus* (*DP-15016*); **E,** *Astragalus hamosus* (*DP-15020*); **F,** *Plantago lanceolata* (*DP-16022*); **G,** *Plantago lanceolata* (*DP-16021*).

PLANTAGINACEAE

Plantago lanceolata L.

▼2*n* = 12, CHN. Bulgaria, Central Rhodope Mts., southward from Parvenetz village, along the ecotrail to the chapel Virgin Mary, on serpentine rocks, 303 m, 42°03′56.28″ N, 24°39′09.43″ E, 15 Aug 2016, *D. Pavlova DP-16022* (SO) [Fig. 5F]; Bulgaria, Eastern Rhodope Mts. near to "Kamennata svatba", Zimzelen village, Kardzali region on calcareous terrains, 309 m, 41°39′22.85″ N, 25°23′57.23″ E, 21 Jul 2016, *D. Pavlova DP-16021* (SO) [Fig. 5G].

The same chromosome number was previously reported for accessions from non-serpentine area in Bulgaria (Kozuharov & Petrova, 1974; Petrova & Stoyanova 1997). The karyotype of the material studied from serpentine consists of eight metacentric, two submetacentric chromosomes with satellites and two subtelocentric chromosomes (Fig. 5F) confirming the karyotype formula presented by Petrova & Stoyanova (1997). The karyotype of the second studied population is symmetrical and consists of metacentric and submetacentric chromosomes without satellites (Fig. 5G).

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* First chromosome count for the taxon.
** New cytotype (chromosome number) for the species.

ARACEAE

Aglaonema commutatum Schott
2*n* = 120, CHN. Brazil, Pernambuco, Recife, cultivated, 21 Jul 1991, *F.C. Ramalho 766-A* (PEUFR).

Alocasia macrorrhizos (L.) G.Don
2*n* = 28, CHN. Brazil, Pernambuco, Recife, Parque de Dois Irmãos, 6 Jun 1991, *F.C. Ramalho 42* (PEUFR) [Fig. 6A].

Anthurium affine Schott
2*n* = 30, CHN. Brazil, Pernambuco, Tapacurá, Reserva Ecológica de Tapacurá, 8 Mar 1991, *F.C. Ramalho 765* (PEUFR); Brazil, Pernambuco, Caruaru, 21 Oct 1991, *F.C. Ramalho 832* (PEUFR) [Fig. 6B].

**Anthurium bromelicola* Mayo & L.P.Felix
2*n* = 30, CHN. Brazil, Pernambuco, Bezerros, Serra Negra, 17 Feb 1997, *S.J. Mayo & L.P. Felix 1156* (EAN) [Fig. 6C].

Anthurium gracile (Rudge) Schott
2*n* = 40, CHN. Brazil, Pernambuco, Recife, Mata de Dois Irmãos, 12 Jul 1991, *F.C. Ramalho 744 & M. Guerra* (PEUFR); Brazil, Pernambuco, Gravatá, Serra das Russas, 10 Jun 2009, *L.P. Felix 12964* (EAN) [Fig. 6D].

Anthurium jilekii Schott
2*n* = 30, CHN. Brazil, Pernambuco, Taquaritinga do Norte, 7 Jan 1993, *M.A. Nadruz & al. 874* (PEUFR) [Fig. 6E].

Anthurium pentaphyllum (Aubl.) G.Don
2*n* = 30 CHN. Brazil, Paraíba, Mamanguape, Cachoeirinha, Pindobal, 30 Sep 2011, *L.P. Felix 13663* (EAN) [Fig. 6F].
2*n* = 60, CHN. Brazil, Pernambuco, Caruaru, Brejo dos Cavalos, 16 Nov 1994, *S. Mayo & al. 907* (PEUFR) [Fig. 6H].

**Anthurium petrophilum* K.Krause
2*n* = 30, CHN. Brazil, Pernambuco, Brejo da Madre Deus, Fazenda Bituri Grande, Lajedo do Cassange, 21 Oct 1992, *F.C Ramalho 13* (PEUFR); Brazil, Paraíba, Esperança, Lagoa de Pedra, 1 Sep 1993, *L.P. Felix 6174* (EAN) [Fig. 6I].

Anthurium scandens (Aubl.) Engl.
2*n* = 48, CHN. Brazil, Pernambuco, Brejo da Madre de Deus, Fazenda Bituri Grande, Mata do Macuco, 21 Dec 1992, *F.C. Ramalho 22* (PEUFR).

**Asterostigma riedelianum* (Schott) Kuntze
2*n* = 34, CHN. Brazil, Pernambuco, Brejo dos Cavalos, 16 Nov 1994, *S. Mayo 1044* (UFP) [Fig. 6G].

Colocasia esculenta (L.) Schott
2*n* = 28, CHN. Brazil, Pernambuco, Recife, cultivated, *M. Guerra 1043* (UFP) [Fig. 6J].

Dieffenbachia seguine (Jacq.) Schott
2*n* = 34, CHN. Brazil, Pernambuco, Timbaúba, Engenho Água Azul, 17 Jul 1992, *F.C. Ramalho 04* (PEUFR).

Dracontioides desciscens (Schott) Engl.
2*n* = 26, CHN. Brazil, Sergipe, Parque Nacional Serra de Itabaiana, 12 Jun 2009, *L.P. Felix 12933* (EAN) [Fig. 7D].

**Dracontium nivosum* (Lem.) G.H.Zhu
2*n* = 26, CHN. Brazil, Pará, Barcarena, Ilha de Trambioca, 10 Jan 2009, *L.P. Felix 12682* (EAN) [Fig. 7A].

**Monstera adansonii* subsp. *klotzschiana* (Schott) Mayo & I.M.Andrade

2*n* = 60, CHN. Brazil, Paraíba, Rebio Guaribas, Mata do Maracujá, 30 Sep 2011, *L.P. Felix 13679* (EAN) [Fig. 7C].

**Montrichardia linifera* (Arruda) Schott

2*n* = 48, CHN. Brazil, Recife, Casa Forte, 10 Jan 1993, *F.C. Ramalho & M. Guerra 20* (PEUFR) [Fig. 7B].

Philodendron acutatum Schott

2*n* = 32, CHN. Brazil, Pernambuco, Recife, Mata de Dois Irmãos, 12 Jun 1991, *F.C. Ramalho 30* (PEUFR) [Fig. 7F].

Philodendron bipinnatifidum Schott ex Endl.

2*n* = 36, CHN. Brazil, Pernambuco, Recife, cultivated, Campus da UFPE, 7 Jun 1991, *F.C. Ramalho 26* (PEUFR) [Fig. 7G].

Philodendron blanchetianum Schott

2*n* = 34, CHN. Brazil, Pernambuco, Cabo de Santo Agostinho, Gurjaú, Mata do Café, 14 Jun 1991, *F.C. Ramalho 27* (PEUFR) [Fig. 7H].

Philodendron fragrantissimum (Hook.) G.Don

2*n* = 46, CHN. Brazil, Pernambuco, Recife, Dois Irmãos, 20 Dec 1992, *F.C. Ramalho 30* (PEUFR) [Fig. 7I].

Philodendron hederaceum (Jacq.) Schott

2*n* = 32, CHN. Brazil, Pernambuco, Cabo, Reserva Ecológica de Gurjaú, Mata do Café, 14 Aug 1991, *F.C. Ramalho 785* (PEUFR) [Fig. 7J].

**Philodendron leal-costae* Mayo & G.M.Barroso

2*n* = 36, CHN. Brazil, Pernambuco, Brejo da Madre de Deus, Fazenda Bituri, 21 Dec 1993, *F.C. Ramalho 11* (PEUFR) [Fig. 7K].

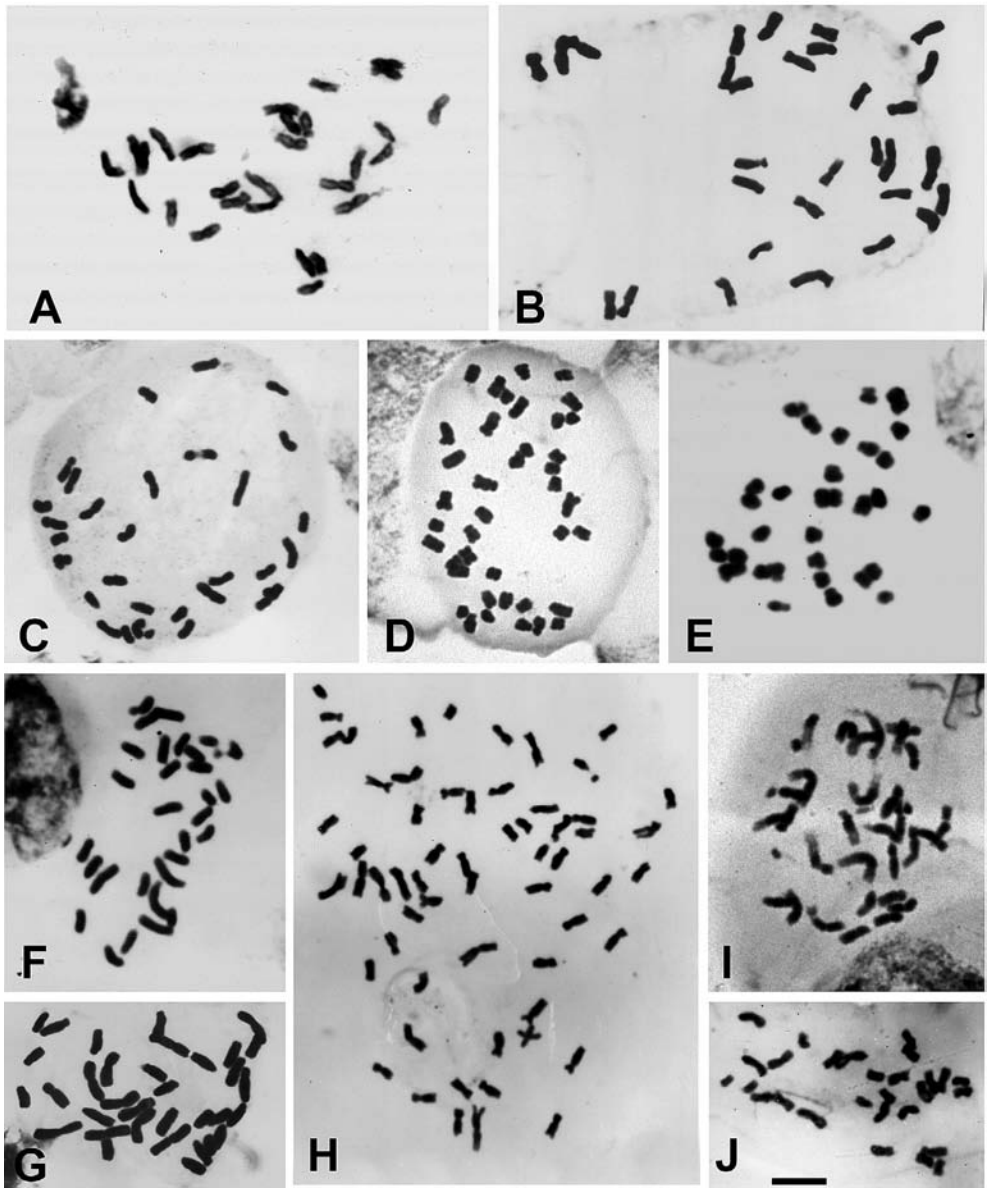

Fig. 6. Mitotic metaphase of: **A,** *Alocasia macrorrhizos*, 2*n* = 28; **B,** *Anthurium affine*, 2*n* = 30; **C,** *Anthurium bromelicola*, 2*n* = 30; **D,** *Anthurium gracile*, 2*n* = 40; **E,** *Anthurium jilekii*, 2*n* = 30; **F,** *Anthurium pentaphyllum*, 2*n* = 30; **G,** *Asterostigma riedelianum*, 2*n* = 34; **H,** *Anthurium pentaphyllum*, 2*n* = 60; **I,** *Anthurium petrophilum*, 2*n* = 30; **J,** *Colocasia esculenta*, 2*n* = 28. — Scale bar = 10 µm

Philodendron ornatum Schott
2*n* = 34, CHN. Brazil, Pernambuco, Timbaúba, Engenho Água Azul, 17 Jul 1992, *F.C. Ramalho 02* (PEUFR) [Fig. 7L].

Philodendron pedatum (Hook.) Kunth
2*n* = 32, CHN. Brazil, Pernambuco, Reserva Ecológica do Tapacurá, 14 Nov 1991, *F.C. Ramalho 834* (PEUFR) [Fig. 8A].

* *Philodendron rudgeanum* Schott
2*n* = 40, CHN. Brazil, Pernambuco, Timbaúba, Engenho Água Azul, 17 Jun 1992, *F.C. Ramalho 01* (PEUFR) [Fig. 8B].

* *Philodendron ruthianum* Nadruz
2*n* = 32, CHN. Brazil, Pernambuco, Cabo de Santo Agostinho, Gurjaú, 14 Jun 1991, *F.C. Ramalho 25* (PEUFR) [Fig. 7E].

Pistia stratiotes L.
2*n* = 28, CHN. Brazil, Paraíba, near the city of Juarez Távora, 15 Aug 2005, *L.P. Felix 10766* (EAN) [Fig. 8C].

Spathicarpa hastifolia Hook.
2*n* = 34, CHN. Brazil, Paraíba, Areia, cultivated, 5 Mar 2013, *L.P. Felix 14854 15* (EAN) [Fig. 8D].

Syngonium podophyllum Schott
2*n* = 26, CHN. Brazil, Pernambuco, Cabo, Reserva Ecológica de Gurjaú, 13 Jan 1993, *M.A. Nadruz & al. 884* (PEUFR) [Fig. 8E].

** *Typhonium roxburghii* Schott
2*n* = 65, CHN. Brazil, Pernambuco, Recife, 4 Feb 1992, *M. Guerra 856* (PEUFR) [Fig. 8F].

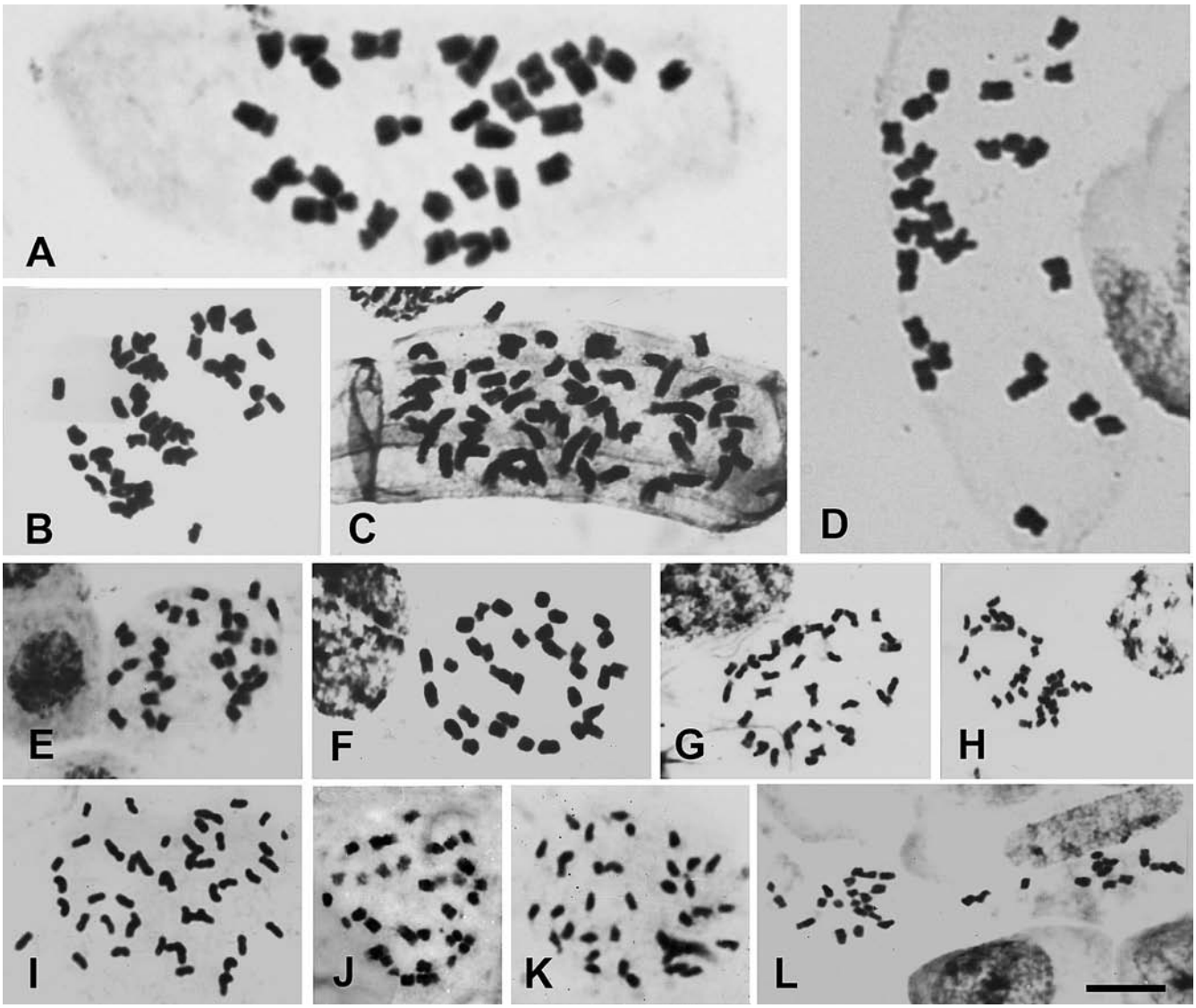

Fig. 7. Mitotic metaphase of: **A,** *Dracontium nivosum*, 2*n* = 26; **B,** *Montrichardia linifera*, 2*n* = 48; **C,** *Monstera adansonii* subsp. *klotzschiana*, 2*n* = 60; **D,** *Dracontioides desciscens*, 2*n* = 26; **E,** *Philodendron ruthianum*, 2*n* = 32; **F,** *Philodendron acutatum*, 2*n* = 32; **G,** *Philodendron bipinnatifidum*, 2*n* = 36; **H,** *Philodendron blanchetianum*, 2*n* = 34; **I,** *Philodendron fragrantissimum*, 2*n* = 46; **J,** *Philodendron hederaceum*, 2*n* = 32; **K,** *Philodendron leal-costae*, 2*n* = 36; **L,** *Philodendron ornatum*, 2*n* = 34. — Scale bar = 10 µm.

** *Xanthosoma sagittifolium* (L.) Schott

2*n* = 39, CHN. Brazil, Pernambuco, UFRPE, 25 Jul 1995, *M. Guerra 1121* (UFP) [Fig. 8G].

Zomicarpa pythonium (Mart.) Schott

2*n* = 20, CHN. Brazil, Pernambuco, Cabo de Santo Agostinho, Praia de Calhetas, 11 Jun 1993, *M. Guerra 927* (UFP) [Fig. 8H].

The family Araceae is a large group of monocots mainly distributed in the Neotropics well represented in Brazil. Regarding chromosome numbers, it is one of the best investigated Neotropical families of angiosperms with karyotype data for more than ¼ of its species (Cusimano & al., 2012). The family is karyologically highly diversified, varying not only in chromosome numbers but also in chromosome size and morphology, distribution of heterochromatin, number and position of rDNA and telomeric sites, ploidy level, sex chromosomes, nuclear DNA amount, etc. (see Lakshmanan & al., 2015; Souza & Renner, 2015). Different chromosome numbers have also been reported for several species, sometimes due to miscounts or taxonomical misidentification (see, e.g., Correia-da-Silva & al., 2014). The putative basic chromosome number of the family has been recently revised and seems to be higher (x = 17) than previously assumed (Cusimano & al., 2012). Aiming to contribute to a better understanding of the cytotaxonomy of the family we report here the chromosome numbers observed in 32 species, including representatives of 17 genera (Table 2).

All samples were collected in the northeast and north of Brazil, including some cultivated ones. The cytological analysis was done using conventional staining with Giemsa (Guerra, 1983). Pictures of most of these species are presented in Figs. 6–8 aiming to illustrate the karyotype variation observed. The highest and the lowest chromosome number were observed in *Aglaonema commutatum* (2*n* = 120) and *Zomicarpa pythonium* (2*n* = 20), respectively. More than half of the species analysed belonged to only two genera: *Anthurium* (7) and *Philodendron* (10).

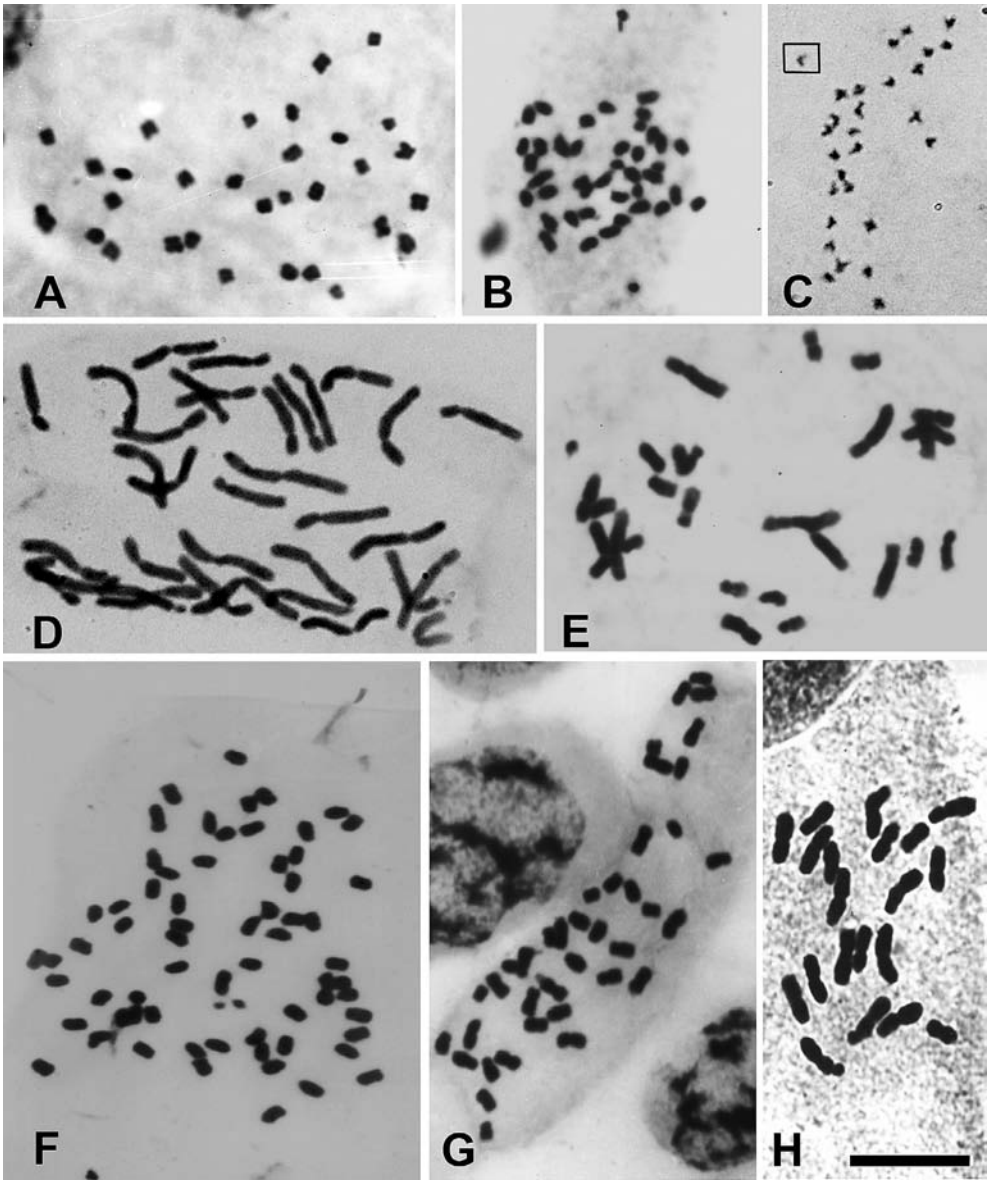

Fig. 8. Mitotic metaphase of: **A,** *Philodendron pedatum*, 2*n* = 32; **B,** *Philodendron rudgeanum*, 2*n* = 40; **C,** *Pistia stratiotes*, 2*n* = 28, distant chromosome highlighted by rectangle; **D,** *Spathicarpa hastifolia*, 2*n* = 34; **E,** *Syngonium podophyllum*, 2*n* = 26; **F,** *Typhonium roxburghii*, 2*n* = 65; **G,** *Xanthosoma sagittifolium*, 2*n* = 39; **H,** *Zomicarpa pythonium*, 2*n* = 20. — Scale bar = 10 µm.

Table 2. List of species analyzed with respective provenances, collector and coll. numbers, chromosome numbers (2*n* and/or *n*), previous counts and sources. Suprageneric clades are organized according to Cusimano & al. (2011). CS14 = Correia-da-Silva & al. (2014); CCDB = Chromosome Counts Database; PA = Pará State; PB = Paraíba State; PE = Pernambuco State; SE = Sergipe State.

Taxon	Provenance	Collector	2*n*	Previous counts (ordered by frequency of counts)		Sources
				n	2*n*	
***Rhaphidophora* clade**						
Monstera adansonii subsp. *klotzschiana* (Schott) Mayo & I.M.Andrade	Rio Tinto, PB	*L.P. Felix 13679*	60			
Lasioideae						
Dracontioides desciscens (Schott) Engl.	Itabaiana, SE	*L.P. Felix 12933*	26		26	CCDB
Dracontium nivosum (Lem.) G.H.Zhu	Barcarena, PA	*L.P. Felix 12682*	26			
Aglaonemateae						
Aglaonema commutatum Schott	Recife, PE	*F.C. Ramalho 766-A*	120		120, 14–(118–120)	CCDB
***Philodendron* clade**						
Philodendron acutatum Schott	Recife, PE	*F.C. Ramalho 30*	32		32, 34, 17	CS14
Philodendron bipinnatifidum Schott ex Endl.	Recife, PE	*F.C. Ramalho 26*	36	18	32, 36, 34–48	CS14
Philodendron blanchetianum Schott	Cabo de Santo Agostinho, PE	*F.C. Ramalho 27*	34		34	CS14
Philodendron fragrantissimum (Hook.) G.Don	Recife, PE	*F.C. Ramalho 30*	46		32	CS14
Philodendron hederaceum (Jacq.) Schott	Cabo, PE	*F.C. Ramalho 785*	32		32, 30, 36	CS14
Philodendron leal-costae Mayo & G.M.Barroso	Brejo da Madre de Deus, PE	*F.C. Ramalho 11*	36			
Philodendron ornatum Schott	Timbaúba, PE	*F.C. Ramalho 02*	34		34	CS14
Philodendron pedatum (Hook.) Kunth	Cabo de Santo Agostinho, PE	*F.C. Ramalho 834*	32		32	CS14
Philodendron rudgeanum Schott	Timbaúba, PE	*F.C. Ramalho 01*	40		32	CS14
Philodendron ruthianum Nadruz	Cabo de Santo Agostinho, PE	*F.C. Ramalho 25*	32			
Spathicarpeae						
Asterostigma riedelianum (Schott) Kuntze	Caruaru, PE	*S. Mayo 1044*	34			
Spathicarpa hastifolia Hook	Areia, PB	*L.P. Felix 14854*	34			
Dieffenbachia seguine (Jacq.) Schott	Timbaúba, PE	*F.C. Ramalho 04*	34	17	34, 36, 68, 40–16, 126–56	CCDB
Tribe Caladieae						
Syngonium podophyllum Schott	Cabo, PE	*M.A. Nadruz & al. 884*	26		26, 24	CCDB
Xanthosoma sagittifolium (L.) Schott	Recife, PE	*M. Guerra 1121*	39	13	26, 24, 28–38	CCDB
Zomicarpa pythonium (Mart.) Schott	Caruaru, PE	*M. Guerra 927*	20		20	CCDB
***Colocasia* clade (20)**						
Colocasia esculenta (L.) Schott	Recife, PE	*M. Guerra 1043*	28	14	28, 42, 38–36, 84, 48, 30–44–52–58–116	
Tribe Areae						
Typhonium roxburghii Schott	Recife, PE	*M. Guerra, 865*	65	26	52, 16–16–65	CCDB

Pothoideae						
Anthurium affine Schott	São Lourenço da Mata, PE	*F.C. Ramalho 765*	30	30		CCDB
	Caruaru, PE	*F.C. Ramalho 832*	30			
Anthurium bromelicola Mayo & L.P.Felix	Bezerros, PE	*S.J. Mayo & L.P. Felix 1156*	30			
Anthurium gracile (Rudge) Schott	Recife, PE	*F.C. Ramalho & M. Guerra 744*	40	40, 30, 20, 60–49		CCDB
	Gravatá, PE	*L.P. Felix 12964*	40			
Anthurium jilekii Schott	Taquaritinga do Norte, PE	*M.A. Nadruz & al. 874*	40			
Anthurium pentaphyllum (Aubl.) G.Don	Mamanguape, PB	*L.P. Felix 13663*	30	15	60, 60+1B	CCDB
	Caruaru, PE	*S. Mayo & al. 907*	60			
Anthurium petrophilum K.Krause	Brejo da Madre de Deus, PE	*F.C. Ramalho 13*	30			
	Esperança, PB	*L.P. Felix 6174*	30			
Anthurium scandens (Aubl.) Engl.	Brejo da Madre de Deus, PE	*F.C. Ramalho 22*	48	16, 24	48, 84, 24, (45–47)	CCDB
***Alocasia* clade**						
Alocasia macrorrhizos (L.) G.Don	Recife, PE		28	14, 21	28, 26, 42	CCDB
***Pistia* clade**						
Pistia stratiotes L.	Juarez Távora, PB	*L.P. Felix 10766*	28	7, 14	28, 14	CCDB
***Aroideae* clade**						
Montrichardia linifera (Arruda) Schott	Recife, PE	*F.C. Ramalho & M. Guerra 20*	48			

For 7 of the 32 species investigated (*Anthurium bromelicola*, *Anthurium petrophilum*, *Asterostigma riedelianum*, *Dracontium nivosum*, *Montrichardia linifera*, *Philodendron leal-costae*, *Philodendron ruthianum*) we did not find any previous chromosome record (Table 2). For *Monstera adansonii* subsp. *klotzschiana* the chromosome number $2n = 60$ was already known but not for this subspecies. For the other 27 species the chromosome numbers reported here are in accordance with at least one of the previous counts, except for *Xanthosoma sagittifolium*, for which we report here a new cytotype with $2n = 39$. According to the Chromosome Counts Data Base (see Rice & al., 2015) several works reported $2n = 26$ for this species, beside a few rare numbers ($2n = 24, 28, 38$). Because of its agronomic importance, protocol for inducing polyploidy has recently been developed but without the expected success (Oumar & al., 2011). The sample here reported is a stable triploid that could be useful in plant breeding programs. The sample was collected on a trail behind the Federal Rural University of Pernambuco, in Recife, apparently escaped from cultivation by small farmers.

The introduced Asiatic species *Typhonium roxburghii* showed $2n = 65$, confirming a previous count by Ramachandran (1978), as *T. divaricatum* Blume. This sample is a pentaploid cytotype, since $2n = 26$ has already been reported for other closely related species and a tetraploid cytotype was also reported by Ramachandran (1978) (see also Souza & Renner, 2015; for a cytotaxonomical review of the genus). The plants analyzed here were collected in a public garden growing as a weed. They were initially cultivated in a single pot in our experimental garden, but in a few years they spread through the whole garden. In spite of the regular flowering, fruits were never observed, possibly due to meiotic sterility caused by pentaploidy. Curiously, we did not find any kind of propagules that could explain its invasive habit.

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* First molecular cytogenetic studies in *C. parvifolium* Sendtn.

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SOLANACEAE

**Capsicum parvifolium* Sendtn.

2*n* = 24, CHN. Brasil, Paraíba: Maturéia, Pico do Jabre, 30 Mar 2009, 07°15′07.99″ S, 37°23′00.99″ W, 1157 m, *F.A. Agra & G.E. Barboza 7075* (JPB).

Capsicum parvifolium is a wild enigmatic chili pepper species from South America. It has an altitudinally discontinuous distribution, being common in areas called "inselbergs" from Brazilian Caatinga (Bahia, Minas Gerais, Paraíba, Ceará, Pernambuco, Piauí, Río Grande do Norte) at 600–1200 m. In Venezuela and Colombia, it occurs at lower altitudes from up to 350 m. It grows as shrub and has stellate flowers with purple lobes, cream tube and white margins (Barboza & al., 2011).

This species has been confused with *C. caatingae* Barboza & Agra due to their similarities in habit, size, shape and color of the corolla and fruit, and with *C. rhomboideum* (Dunal) Kuntze, due to their similarities in the calyx. The latter, however, can be easily differentiated by corolla shape and color (Barboza & al., 2011).

Mitotic chromosomes were observed in squashed root meristems. Root tips were pretreated with p-dichlorobenzene saturated solution for 2 h at room temperature, fixed in absolute ethanol/glacial acetic acid (3 : 1 v/v) for 12 h and stored at −20°C. Root tips were digested in a 2% cellulase and 20% pectinase solution at 37°C for 30 min. The constitutive heterochromatin was analyzed using fluorescence banding (CMA/DA/DAPI), according to Schweizer & Ambros (1994), and the ribosomal loci (rDNA) using fluorescent in situ hybridization (FISH), following the protocol of Schwarzacher & Heslop-Harrison (2000), with modifications. For details of elaboration and labelling of rDNA probes see Romero & al. (2015). For karyotype description, six metaphase plates were measured and chromosomes were arranged in groups according to centromere position and in decreasing order of size within each type. Chromosome terminology follows Levan & al. (1964). Satellite length was excluded for calculation of the arm ratio of *sm* chromosomes.

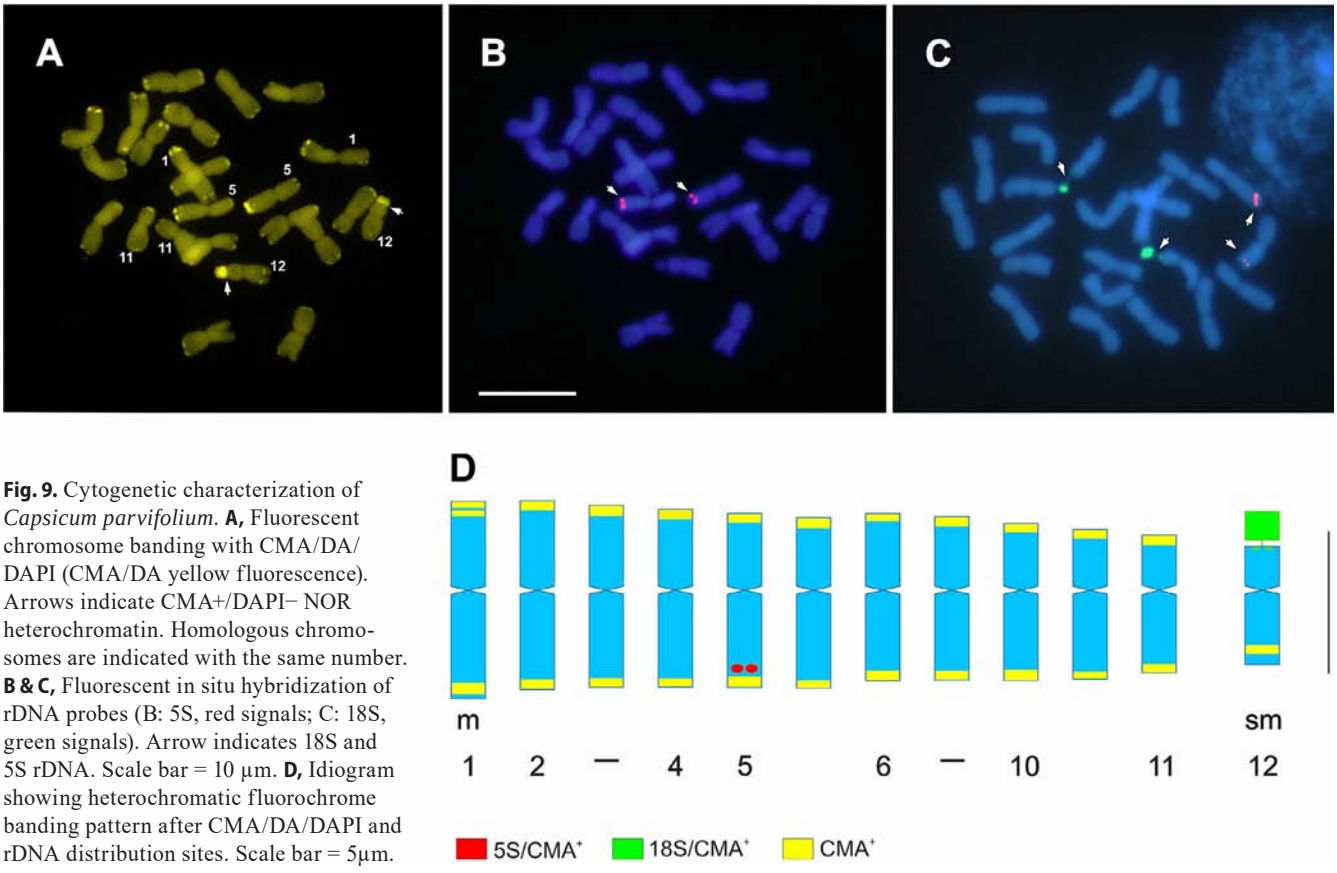

Fig. 9. Cytogenetic characterization of *Capsicum parvifolium*. **A,** Fluorescent chromosome banding with CMA/DA/DAPI (CMA/DA yellow fluorescence). Arrows indicate CMA+/DAPI− NOR heterochromatin. Homologous chromosomes are indicated with the same number. **B & C,** Fluorescent in situ hybridization of rDNA probes (B: 5S, red signals; C: 18S, green signals). Arrow indicates 18S and 5S rDNA. Scale bar = 10 µm. **D,** Idiogram showing heterochromatic fluorochrome banding pattern after CMA/DA/DAPI and rDNA distribution sites. Scale bar = 5µm.

Capsicum parvifolium showed karyotype formulae with 11*m*+1*sm* chromosomes and haploid karyotype length of 71.11 µm. The short arm of pair 12 (*sm*) carries a satellite (Fig. 9A–D). All chromosome pairs presented GC-rich regions at the chromosome ends of both arms, including the satellite of pair #12 (Fig 12A).

The 5S rDNA probe showed two strong hybridization signals in interstitial region at a long arm of one metacentric chromosome pair, which is consistent with the intercalary CMA+ band (Fig. 9A–C). FISH with 18S rDNA probe showed two signals in a submetacentric chromosome pair (#12) (Fig. 9C).

The plant material studied by Moscone (1993) and Moscone & al. (1993, 1995, 2007) as *C. parvifolium* has been recognized as *C. caatingae* (Barboza & al., 2011). Nevertheless, the heterochromatic pattern of *C. parvifolium* resembles those found in *Capsicum* species endemic to Caatinga biome (unpub. data), especially that of *C. caatingae* (cytotype #2) (Moscone & al., 2007).

Moscone & al. (2003) observed a strong correlation between genome size and heterochromatin amount in *Capsicum* species, and Scaldaferro & al. (2013) found the same correlation between heterochromatin amount and haploid karyotype length. *Capsicum parvifolium* has a lower heterochromatin amount than species of *Capsicum* of the same genome size (data not shown), such as *C. campylopodium* Sendtn. (Moscone & al., 2003). For *Capsicum*, Moscone & al. (2003) proposed increases not only in highly tandem repetitive sequences of heterochromatic regions but also in dispersed DNA repeats.

The FISH showed a conserved 5S rDNA signal number in all *Capsicum* species studied (Romero, 2013; Romero & al., 2015; Scaldaferro & al., 2016). However, this is the first report of 5S rDNA located on the long arm within the genus. The 18S rDNA FISH pattern found is different from the fluorescence banding observed, which is in contrast to previous records of other *Capsicum* species (Romero, 2013; Romero & al., 2015; Scaldaferro 2010).

Capsicum parvifolium shares chromosome characteristics with two groups of *Capsicum* species; the $x = 13$ group and the group endemic to Caatinga. The presence of one pair of nucleolar organizer regions (NORs) is shared with Brazilian and Venezuelan $x = 13$ *Capsicum* species; and heterochromatin pattern and karyotype formulae are similar to those of Caatinga *Capsicum* species. A recent phylogenetic study showed a close relationship between *C. parvifolium* and *C. caatingae* (Barboza, pers. comm.).

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