



Is there a relationship between polyploidy and stressful environments? A case study of inselbergs in northeastern Brazil

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ABSTRACT. The aim of this study was to examine the hypothesis that plants with higher ploidy levels are selected by environments under more extremely stressful conditions than the same (or closely related) species from less rigorous terrestrial or epiphytic habitats. Therefore, we analyzed the chromosome numbers of 26 species belonging to 21 genera and 13 families of angiosperms that were collected on 4 inselbergs in Paraíba State, northeastern Brazil. In addition, 13 rupicolous species that grow in the study area, whose chromosome numbers were previously determined, were included in our analysis. Plant chromosome numbers varied between $2n = 12$ in *Callisia filiformis* (Commelinaceae) to $2n = \text{ca. } 240$ in *Epidendrum cinnabarinum* (Orchidaceae). The data as a whole do not support the hypothesis that inselberg species have greater levels of ploidy than terrestrial or epiphytic populations of the same or closely related

species because these rupicolous species demonstrate chromosomal evolution patterns that are independent of the stressful environments of inselberg formations.

Key words: Chromosome number variation; Karyotype; Cytogenetics; Cytotypes; Chromosome evolution; Whole-genome duplication

INTRODUCTION

Karyotype analyses are an important tool for evolutionary studies and can reveal different strategies of chromosomal evolution (Guerra, 2012). In these analyses, chromosome number is one of the most used, reliable, and practical features for characterizing a large number of plant species. Different from morphological or biochemical characteristics, which depend on the expression of some genes, chromosomal characteristics have the great advantage that the karyotype is nothing less than the genome itself and that its phenotype at metaphase does not depend on gene expression, environmental conditions, age, and developmental phase (Guerra, 2012).

An important and particularly widespread feature in the phylogeny of flowering plants is polyploidy (whole-genome duplication), which is considered to be the major driving force behind the chromosome number variation in angiosperms. This mechanism is very important for speciation, and it has profound impacts on biodiversity dynamics and ecosystem functioning (Ainouche and Jenczewski, 2010). Molecular analyses suggest that the genomes of 90% extant angiosperms retain evidence of one or more ancient genome-wide duplications (Leitch and Leitch, 2008). Despite the occurrence of polyploidy in angiosperms, its implications to evolution have long been debated (Leitch and Leitch, 2008; McIntyre, 2012). In the face of this constant occurrence, some questions arise about the advantages of polyploidy, such as greater abiotic and biotic tolerances (McIntyre, 2012). In orchids, the occurrence of terrestrial or rupicolous growth habitats among species belonging to typically epiphytic genera is often correlated with polyploidy. Many rupicolous species of the genus *Cattleya*, for example, are tetraploids, while the epiphytic species are exclusively diploids. Other genera of orchids, such as *Oncidium* and *Epidendrum* (Felix and Guerra, 2010), are also polyploid when rupicolous, suggesting that the occurrence of these cytotypes is related in some way to the occupation of these habitats.

Many rupicolous species are found in inselbergs (from German insel: island and berg: mountain), that are rock outcrops, which are frequently huge, giga-year-old monoliths that mainly consist of Precambrian granites or gneisses and are defined as remnants of erosion processes within a plain landscape (Sarhou et al., 2010). This single model of environment supports ecologically isolated vegetations that are quite distinct from those of the surrounding area because of the extreme edaphic and climatic conditions that are encountered on these rock faces (Parmentier et al., 2005). Species that inhabit inselbergs are extremely well adapted to water stress, high solar radiation, high temperatures, and high levels of evapotranspiration because of evolutionary sympatric differentiation in relation to the species around them (Parmentier et al., 2005). Inselbergs host flora that originated from ancestral plants and were probably derived from more stable terrestrial or epiphytic environments, resulting in a spatial pattern of species that was promoted by stochastic processes,

such as dispersal and demographic drift, which can lead to colonization and local extinction, respectively (Parmentier et al., 2005).

The notion of the adaptive value of polyploidy has its origin in the observation that highly polymorphic species form polyploid complexes adapted to extreme habitats with strong environmental pressure, such as *Galium anisophyllum* (alpine areas) and *Cruciata coronata* (dry environments) (Ehrendorfer, 1970). The increased adaptability of polyploids in relation to their diploid progenitors has been confirmed in artificial neo-polyploids, where the reduction in both pollen and seed fertility in the F1 generation is rapidly recovered in later generations (Ramsey and Schemske, 2002). Positive genetic combinations derived from polyploid events could result in an increased capacity of these polyploids to colonize habitats that were previously unusable by their diploid progenitors (Soltis et al., 2004). Most orchid species are epiphytic with terrestrial ancestors (Benzing, 1990), and most of these epiphytes are hexaploids (Felix and Guerra, 2005). Some groups within the family Orchidaceae that grow on inselbergs have a distinctly higher ploidy level than related epiphytic species (Felix and Guerra, 2010). To test the hypothesis that plants growing under stressful conditions tend to have higher ploidy levels, we examined the chromosome number of 26 species belonging to 13 families of angiosperms that occur on various inselbergs in northeastern Brazil. These species were among the most representative of this flora in a previous floristic survey of inselbergs in this region (Pitrez, 2006). In order to increase the sample size, we included an additional 13 inselberg species with previous chromosomal reports.

MATERIAL AND METHODS

Plant material

All the species examined were collected on inselbergs in Paraíba State, Brazil, especially on 4 peaks in the municipalities of Esperança, Fagundes, Pocinhos, and Serraria. In order to identify the best-represented groups on these inselbergs, floristic and phytosociological studies were first undertaken (Pitrez, 2006). In addition to the species collected on these 4 outcrops, 2 species (*Rauwolfia ligustrina* and *Cissus sulcicaulis*) were collected from inselbergs in the municipalities of Araruna and Barra de Santa Rosa (also in Paraíba), respectively. Material collected was cultivated in the experimental garden at the Departamento de Fitotecnia of Universidade Federal da Paraíba before cytogenetic examination. Reference material was deposited in the Jayme Coelho de Moraes Herbarium (EAN) of Centro de Ciências Agrárias of Universidade Federal da Paraíba.

Slide preparation

Root tips were pre-treated with 0.002 M 8-hydroxyquinoline Sigma-Aldrich (Saint Louis, MO, USA) for 4 to 24 h at 4°C, fixed in Carnoy's solution (3:1 absolute ethanol:acetic acid, v/v - Merck KGaA, Darmstadt, Germany) for a period of 3 to 24 h at room temperature, and stored at -20°C for later analysis. For slide preparation, root tips and flower buds were washed in distilled water to remove the fixative, hydrolyzed in 5 N HCl (Merck KGaA) for 20 min at 20°C, and washed in distilled water.

Squash preparations were made in a drop of 45% acetic acid (Merck KGaA). After

the cover slips were removed by immersion in liquid nitrogen, they were allowed to dry at room temperature, stained with 2% Giemsa (Merck KGaA) for 10 min, and mounted in Entellan (Merck KGaA). Interphase nuclei and patterns of prophase condensation were classified and photographed using a light microscope BX41 Olympus (Tokyo, Japan) fitted with an Olympus D-540 digital camera.

RESULTS

Twenty-six species belonging to 21 genera and 13 families of angiosperms were collected on the inselbergs and studied. Eighteen of these species (*Alstroemeria longistaminea*, *Hippeastrum stylosum*, *Mandevilla tenuifolia*, *Rauwolfia ligustrina*, *Aristolochia birostris*, *Eupatorium ballotaefolium*, *Melocactus bahiensis*, *Melocactus ernestii*, *Pilosocereus pachycladus*, *Clusia nemorosa*, *Tradescantia ambigua*, *Euphorbia comosa*, *Stillingia trapezoidea*, *Paliavana tenuiflora*, *Sinningia nordestina*, *Phyllanthus claussenii*, and *Cissus sulcicaulis*) were karyotyped for the first time. Table 1 lists the species that were examined, their chromosome numbers, collection locations, and data concerning other inselberg species known to occur in the survey area that were previously examined in published (or unpublished) studies. Most species that were examined demonstrated semi-reticulated interphase nuclei with lightly stained chromatin filaments, irregularly shaped chromocenters, and chromosomes with proximal condensation patterns. Only *Alstroemeria* sp, *Hippeastrum stylosum*, and *Tradescantia ambigua* had reticulated interphase nuclei, uniformly distributed chromatin, and a distinctly continuous prophase condensation pattern. The chromosomal counts of the other 12 species were confirmed by at least one of the previously published reports. The only exception was *Eupatorium ballotaefolium*.

Alstroemeria longistaminea, of the family Alstroemeriaceae (Figure 1A), with $2n = 16$, demonstrated a bimodal karyotype with 2 metacentric pairs. The largest pair was $19.3 \mu\text{m}$ long and the shortest was $7.3 \mu\text{m}$. The other chromosomes were acrocentric and gradually diminishing in size, from 9.3 to $6.1 \mu\text{m}$, with a terminal satellite on the short arm of one of the largest acrocentric chromosomes.

Hippeastrum stylosum (Amaryllidaceae) showed $2n = 22$, and chromosomes ranged from 4.6 to $11.3 \mu\text{m}$ (Figure 1B). The karyotype of this species is formed by 4 pairs of small metacentric chromosomes and 7 large submetacentric and acrocentric pairs.

Two species were analyzed from the family Apocynaceae: *Mandevilla tenuifolia* (Figure 1C), with $2n = 20$, and *Rauwolfia ligustrina*, with $2n = 22$ (Figure 1D). The former species has larger chromosomes (1.0 to $1.7 \mu\text{m}$) than the latter (0.3 to $1.3 \mu\text{m}$). In both species, the chromosome morphology was difficult to define. The present count of $2n = 20$ for *Mandevilla tenuifolia* is the first for the genus.

Aristolochia birostris (Aristolochiaceae) demonstrated a chromosome number of $2n = 14$ (Figure 1E), with metacentric and submetacentric chromosomes that were 1.0 to $1.7 \mu\text{m}$.

Eupatorium ballotaefolium (Asteraceae) has $2n = 20$ chromosomes (Figure 1F). This species demonstrated a symmetrical karyotype that was formed by 1 acrocentric chromosomal pair that was 1.2 to $2.1 \mu\text{m}$ long (the other pairs were submetacentric to metacentric), a pair of submetacentric chromosomes with a terminal nucleolus organizer region (NOR), and another metacentric chromosome with a proximal NOR.

Table 1. List of families and species, material examined and their origins, chromosome numbers, and bibliographic sources.

Taxa	Voucher	Provenance	Chromosome number (2n)	Source
Astromeriaceae				
<i>Alstroemeria longistaminea</i> Mart. ex Schult. & Schult. f.	S.Pitrez 392	Fagundes, PB	16	PS
Amaryllidaceae				
<i>Habenanthus itabobinus</i> Ravenna	S.Pitrez 500	Pocinhos, PB	42 + 1B	Felix et al., 2011
<i>Hippeastrum stylosum</i> Herb	S.Pitrez 587	Fagundes, PB	22	PS
Apocynaceae				
<i>Mandevilla tenuifolia</i> (Mikan) R.E. Woodson	S.Pitrez 507	Serraria, PB	20	PS
<i>Rauwolfia ligustrina</i> Willd. ex Roem. & Schult.	S.Pitrez 355	Araruna, PB	22	PS
Aristolochiaceae				
<i>Aristolochia birostis</i> Duch.	A.Almeida 383	Esperança, PB	14	PS
Asteraceae				
<i>Eupatorium ballotaefolium</i> H. B. & K	S.Pitrez 640	Fagundes, PB	20	PS
Bromeliaceae				
<i>Hohenbergia catingae</i> Ule			50	Cotias-de-Oliveira et al., 2000
Cactaceae				
<i>Cereus jamacaru</i> DC.	S.Pitrez S/N	Esperança, PB	22	PS
<i>Melocactus bahiensis</i> (Britton & Rose) Luetzelb.	S.Pitrez S/N	Pocinhos, PB	44	PS
<i>Melocactus ernestii</i> Vaupel	A.Almeida 240	Esperança, PB	44	PS
<i>Pilosocereus pachycladus</i> Ritter	S.Pitrez S/N	Pocinhos, PB	44	PS
Clusiaceae				
<i>Clusia nemomosa</i> G. F. W. Mey.	S.Pitrez S/N	Araruna, PB	60	PS
<i>Clusia</i> sp	L.P.Felix S/N		60	PS
Commelinaceae				
<i>Callisia filiformis</i> (Martens & Galeotti) D.R. Hunt		Fernando de Noronha, PE	14	Pitrez, 1998
<i>Callisia repens</i> L.		Bezerros, PE	12	Pitrez, 1998
<i>Commelina erecta</i> L.		Camargibe, PE	60	Pitrez et al., 2001
<i>Commelina obliqua</i> Vahl		Bezerros, PE	60	Pitrez et al., 2001
<i>Tradescantia ambigua</i> Mart.	S.Pitrez 601	Pocinhos, PB	24	PS
Convolvulaceae				
<i>Evolvulus filipes</i> Mart.	L.P.Felix 10202	Teixeira, PB	26	PS; Pitrez et al., 2008
<i>Evolvulus glomeratus</i> Nees & C. Mart.	A.Almeida 405	Pocinhos, PB	26	PS; Pitrez et al., 2008
<i>Ipomoea longiramosa</i> Choisy	A.Almeida 396	Pocinhos, PB	30	PS; Pitrez et al., 2008
<i>Ipomoea marcelia</i> Meisn.	S.Pitrez 250	Pocinhos, PB	30	PS; Pitrez et al., 2008
<i>Ipomoea pikei</i> Hoehne	A.Almeida 411	Araruna, PB	30	PS; Pitrez et al., 2008
<i>Jacquemontia densiflora</i> Hallier. F.	S.Pitrez 409	Pocinhos, PB	18	PS; Pitrez et al., 2008
<i>Merremia aegyptia</i> (L.) Urb.	S.Pitrez 562	Pocinhos, PB	30	PS; Pitrez et al., 2008

Continue on next page

Table 1. Continued

Taxa	Voucher	Provenance	Chromosome number (2n)	Source
Euphorbiaceae				
<i>Euphorbia comosa</i> Vell.	S.Pitrez 446	Esperança, PB	40	PS
<i>Stillingia trapezoides</i> Ule	S.Pitrez 200	Esperança, PB	36	PS
Gesneriaceae				
<i>Paltanova tenuiflora</i> Mansf.	S.Pitrez 379	Fagundes, PB	28	PS
<i>Sinningia nondestina</i> Chateaux & Baracho	A.Almeida 363	Serraria, PB	26	PS
Orchidaceae				
<i>Acianthera ochreate</i> (Lindl.) Pridgeon & M. W. Chase		Bezerros, PE	40	Felix and Guerra, 2010
<i>Brassavola tuberculata</i> Hook.		Camocim de São Felix, PE	40	Felix and Guerra, 2010
<i>Cyrtopodium intermedium</i> Brade		Bezerros, PE	46	Felix and Guerra, 2000
Orchidaceae				
<i>Cyrtopodium polyphyllum</i> (Vell.) Pabst ex F. Barros		Bezerros, PE	46	Felix and Guerra, 2000
<i>Epidendrum cinnabarinum</i> Salzm. ex Lindl.		Camocim de São Felix, PE	Ca. 240	Felix and Guerra, 2010
<i>Epidendrum secundum</i> Jacq.		Camocim de São Felix, PE	68	Felix and Guerra, 2010
<i>Prescoitia phleoides</i> Lindl.		Bezerros, PE	48	Felix and Guerra, 2005
Phyllanthaceae				
<i>Phyllanthus clausenii</i> Müell. Arg.	S.Pitrez 335	Araruna, PB	26	PS
Vitaceae				
<i>Cissus sulcataulis</i> Planch	L.P.Felix S/N	Barra de Santa Rosa, PB	34	PS

PB = Paraíba State, Brazil; PE = Pernambuco State, Brazil; PS = present study.

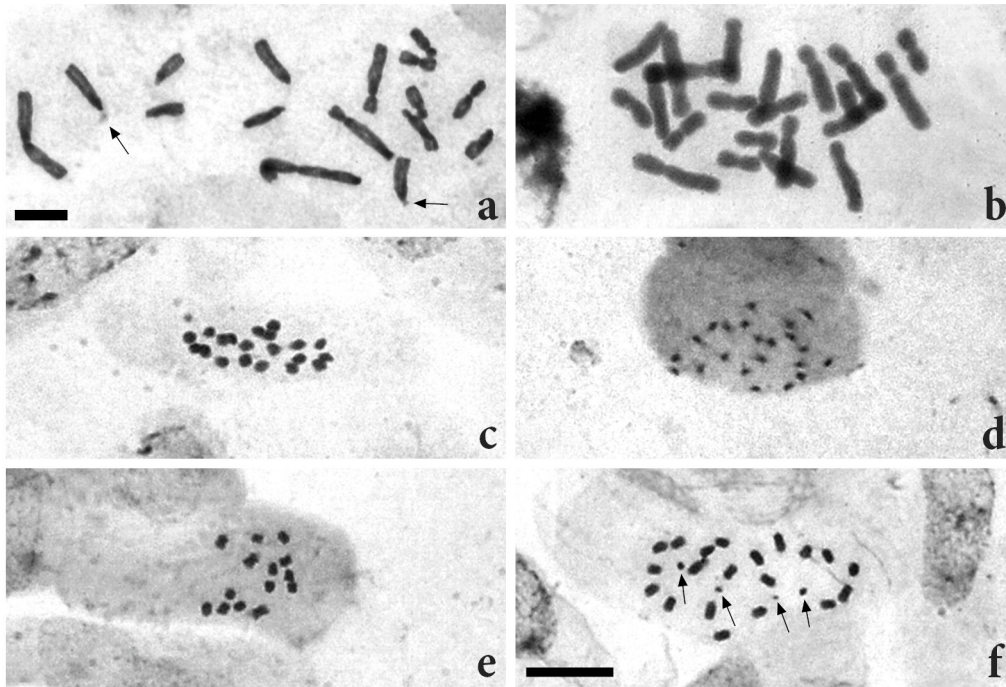


Figure 1. Chromosome complement of: **a.** *Alstroemeria longistaminea* ($2n = 16$); **b.** *Hippeastrum stylosum* ($2n = 22$); **c.** *Mandevilla tenuifolia* ($2n = 20$); **d.** *Rauwolfia ligustrina* ($2n = 22$); **e.** *Aristolochia birostris* ($2n = 14$); **f.** *Eupatorium ballotaefolium* ($2n = 20$). Arrows indicate satellites. Bar in F = 10 μm .

All the species of Cactaceae examined to date demonstrated a symmetrical karyotype with metacentric and submetacentric chromosomes. *Cereus jamacaru* (Figure 2A) presented $2n = 22$. The chromosome number $2n = 44$ and chromosome lengths of 1.3 to 2.6 μm that were registered here for *Pilosocereus pachycladus* (Figure 2B) are the first report of polyploidy for the genus. Both *Melocactus bahiensis* and *Melocactus ernestii* (Figure 2C and D) gave unique counts for the genus, $2n = 44$, and had chromosome lengths ranging from 1.0 to 2.1 μm .

Two species of Clusiaceae were examined here: *Clusia nemorosa* (Figure 2E) and *Clusia* sp (Figure 2F), both with $2n = 60$, a symmetrical karyotype, and metacentric to submetacentric chromosomes.

Tradescantia ambigua (Commelinaceae) (Figure 3A) had $2n = 24$ and a symmetrical karyotype with large metacentric chromosomes with lengths that ranged from 5.0 to 8.7 μm .

Within the genus *Evolvulus* (Convolvulaceae), *Evolvulus filipis* (Figure 3B), and *Evolvulus glomeratus* (Figure 3C) demonstrated symmetrical karyotypes with $2n = 26$; chromosomes were 0.8 to 1.5 μm in the former and 1.3 to 2.1 μm in the latter. Three species of the genus *Ipomoea* (*I. longeramosa*, *I. marcellia*, and *I. pikeli*) demonstrated $2n = 30$ (Figure 3D-F), symmetrical karyotypes, metacentric to submetacentric chromosomes that were 0.3 to 1.6 μm , and satellites in some of the cells of all the 3 species.

The largest chromosomes among the Convolvulaceae were observed in *Jacquemontia densiflora* (Figure 3G), which has $2n = 18$ and a symmetrical karyotype with predominantly submetacentric chromosomes that range from 2.1 to 3.8 μm . *Merremia aegyptia* (Figure 3H) has $2n = 30$, a symmetric karyotype with metacentric to submetacentric chromosomes measuring 1.3 to 2.3 μm , and a secondary terminal constriction on one of the submetacentric pairs.

Euphorbia comosa (Euphorbiaceae) is widely distributed among all of the inselbergs. It had a chromosome number of $2n = 40$ (Figure 4A), and its chromosomes were 1.0 to 3.3 μm . *Stillingia trapezoidea* had $2n = 36$ (Figure 4B), large chromosomes that were 2.0 to 3.5 μm , all of which were apparently acrocentric, and a secondary proximal constriction on one of the largest chromosome pairs.

Two species of the family Gesneriaceae were analyzed, *Paliavana tenuiflora* had $2n = 28$ (Figure 4C), and *Sinningia nordestina* presented $2n = 26$ (Figure 4D). Both of these species demonstrated small chromosomes whose morphology was difficult to define.

Phyllanthus claussenii (Phyllanthaceae) had $2n = 26$ (Figure 4E) and very small chromosomes (0.6 to 1.6 μm). *Cissus sulcicaulis* (Vitaceae) demonstrated $2n = 34$ and small meta-centric to submetacentric chromosomes (Figure 4F).

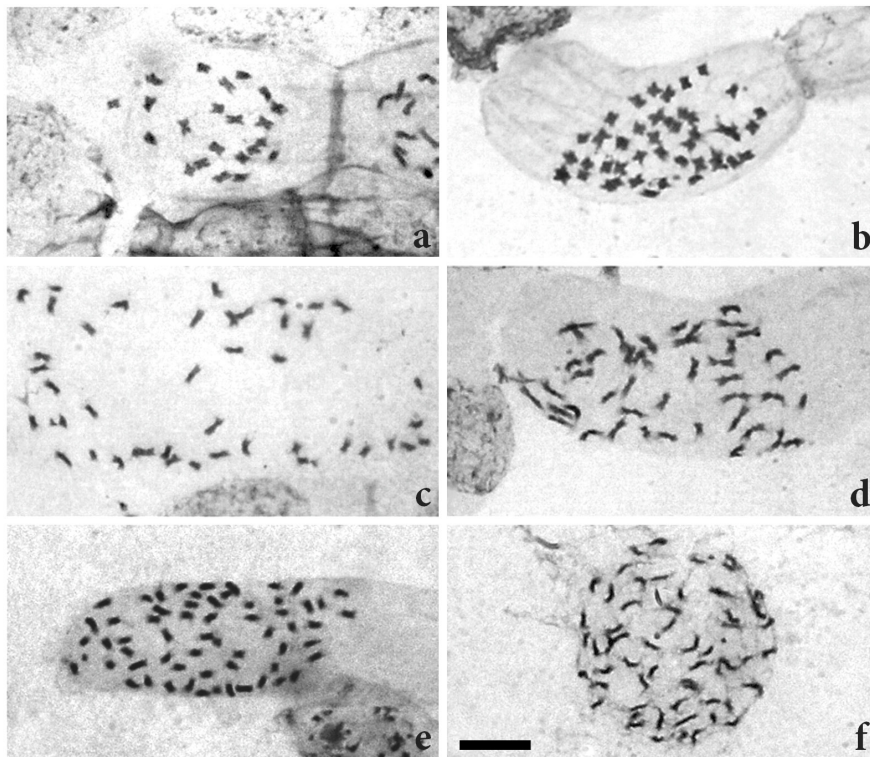


Figure 2. Chromosome complement of: **a.** *Cereus jamacaru* ($2n = 22$); **b.** *Pilosocereus pachicladius* ($2n = 44$); **c.** *Melocactus bahiensis* ($2n = 44$); **d.** *M. ernesti* ($2n = 44$); **e.** *Clusia nemorosa* ($2n = 60$); **f.** *Clusia* sp ($2n = 60$). Bar in F = 10 μm .

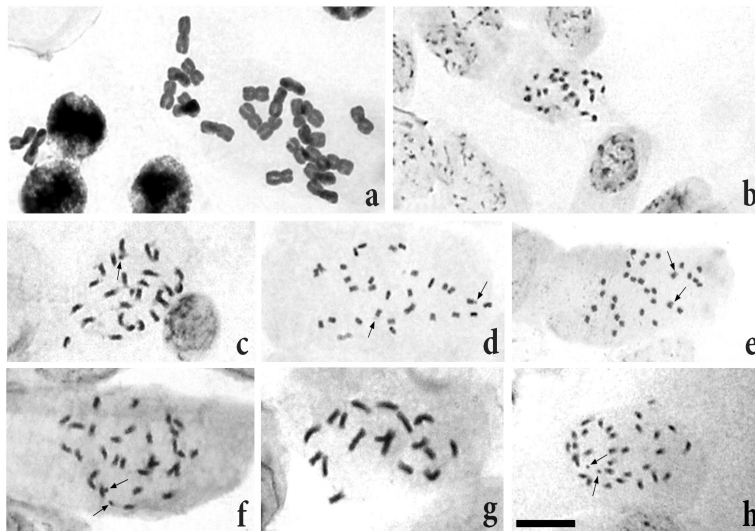


Figure 3. Chromosome complement of: **a.** *Tradescantia ambigua* ($2n = 24$); **b.** *Evolvulus filipis* ($2n = 26$); **c.** *E. glomeratus* ($2n = 26$); **d.** *Ipomoea longeramosa* ($2n = 30$); **e.** *I. marcellia* ($2n = 30$); **f.** *I. pikeli* ($2n = 30$); **g.** *Jacquemontia densiflora* ($2n = 18$); **h.** *Merremia aegyptia* ($2n = 30$). Arrows indicate satellites. Bar in H = 10 μm .

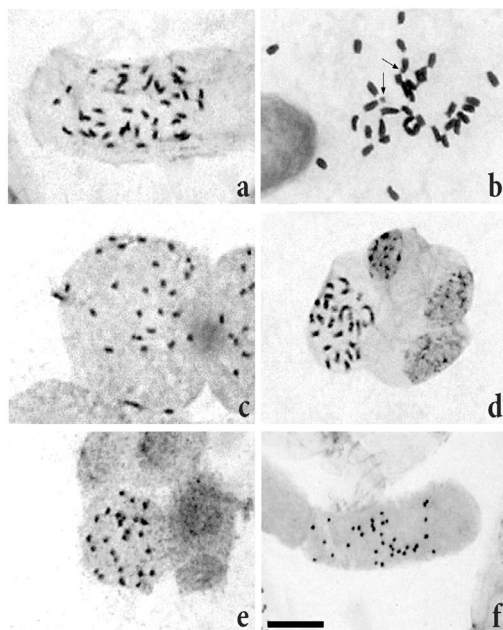


Figure 4. Chromosome complement of: **a.** *Euphorbia comosa* ($2n = 40$); **b.** *Stillingia trapezoidea* ($2n = 36$); **c.** *Paliavana tenuiflora* ($2n = 28$); **d.** *Sinningia nordestina* ($2n = 26$); **e.** *Phyllanthus clausseni* ($2n = 26$); **f.** *Cissus sulcicaulis* ($2n = 34$). Arrows indicate satellites. Bar in F = 10 μm .

DISCUSSION

In addition to the 26 species that were cytogenetically investigated in this study, we included data previously published concerning 13 other species encountered on other inselbergs in northeastern Brazil and were present on the inselbergs examined here (Table 1), expanding the sample to a total of 39 species belonging to 31 genera and 15 families. There is one report of polyploidy in *Commelina erecta* and *Commelina obliqua* of the family Commelinaceae (Pitrez et al., 2001) and in *Epidendrum cinnabarinum* and *Epidendrum secundum* of the family Orchidaceae (Felix and Guerra, 2010). The single species of Bromeliaceae from the inselbergs examined here did not demonstrate different ploidy levels. Other representatives of the families Commelinaceae (3 species) and Orchidaceae (6 species) found on the inselbergs likewise did not demonstrate ploidy levels higher than those observed in related terrestrial species, except *Habenaria obtusa*, which had a single triploid individual (Felix and Guerra, 2005).

A wide range of chromosome numbers was observed among species of Orchidaceae on the inselbergs. Of the species in the area studied with previously published karyotypes, *Brasavola tuberculata* had $2n = 40$, *Cyrtopodium intermedium* and *Cyrtopodium polyphyllum* had $2n = 46$, *Epidendrum cinnabarinum* had $2n = 240$, *Epidendrum secundum* had $2n = 68$, *Habenaria obtusa* had $2n = 50$, *Pleurothallis ochreatea* had $2n = 40$, and *Prescottia phleoides* had $2n = 48$ (Felix and Guerra, 2005, 2010). Of these species, *Habenaria* and *Prescottia* are typically terrestrial or rupicolous, while the other species are usually epiphytic. These chromosome numbers point to differences in ploidy levels only for *Epidendrum secundum* and *Epidendrum cinnabarinum*, while the other species are considered to be paleopolyploids that stabilized at the hexaploid level (Felix and Guerra, 2005) and are considered as diploids. Among the Bromeliaceae, the only species from the area studied that was previously analyzed did not have a chromosome number that differed from the most frequent counts for the family, independent of whether they occurred as epiphytes or in rupicolous or terrestrial environments.

The chromosome number $2n = 22$ observed for *Hippeastrum stylosum* (Amaryllidaceae) was the same as that previously and frequently reported for this species and for others of the same genus (Dutilh, 1989). The karyotype that we observed also coincided with that previously described by Guerra (1986) for a population of the same species from Pernambuco State (also in northeastern Brazil), although those were from a terrestrial environment. Nevertheless, there is one report of polyploidy in *Habranthus itaobinus* (Felix, 2009), and intraspecific polyploidy was frequently observed among terrestrial species of *Zephyranthes*, including *Zephyranthes sylvatica* from Pernambuco State in northeastern Brazil (Felix et al., 2008).

Aristolochia birostris (Aristolochiaceae) with $2n = 14$ is often found in terrestrial habitats. The cytogenetic analysis of this species coincides with the chromosome number that was most frequently reported for the genus (Berjano et al., 2009). A previous chromosomal analysis (Berjano et al., 2009) of material collected in terrestrial environments in the neighboring state of Pernambuco demonstrated essentially the same karyotype as the present sample.

The chromosome number of *Eupatorium ballotaefolium* ($2n = 20$), a weedy Asteraceae that is common on inselbergs and in other terrestrial environments, agrees with counts that are frequently associated with this genus, but it diverged from all numbers previously reported ($2n = 10, 30$) for this species (Moore, 1973). Other weedy and invasive species of Asteraceae in the Caatinga vegetation of the states of Paraíba and Rio Grande do Norte likewise demonstrated variability relative to previously published chromosomal counts, possibly because of

the existence of cryptic species. The taxonomically similar weedy species *Emilia sonchifolia* ($2n = 10$) and *Emilia sagittata* ($2n = 20$) demonstrate the same karyotype variation (Guerra and Nogueira, 1990), although it is not related to climatic stress on the inselbergs.

The chromosome number reported here for *Pilosocereus pachycladus* ($2n = 44$) is the first report of polyploidy for the genus, which had only one other previously published count, $2n = 22$ in *Pilosocereus lanuginosus* (Baker, 2002). The chromosome number in *Cereus jama-caru* ($2n = 22$) confirmed previously published data for this species (Pedrosa et al., 1999) from a population in Pernambuco State. Cacti constitute a taxonomic group that is highly adapted to xeric environments, and they are distributed principally within the American continents. In spite of their adaptations to rigorous and dry environments, the family is predominantly diploid, with $2n = 22$. Examples of intra- and inter-specific polyploidy have been observed in numerous cacti genera, but no obvious correlations between polyploid forms and the occupation of water-stressed habitats were demonstrated. The epiphytic genera of the family, such as *Rhypsalis* and *Phyllocladus* (both epiphytic and found in humid forests) have the same $2n = 22$ count that is most frequently seen in species from desert regions.

The same chromosome number presented here for *Clusia nemorosa* and *Clusia* sp (Clusiaceae) was observed in 4 other species of *Clusia* that were examined by Cruz et al. (1990), indicating that $x = 30$ is the basic number for a majority of the species of the genus and suggesting that it is a paleopolyploid group. Numerous species of *Clusia* occur on inselbergs in humid environments, as terrestrial plants in humid forests and epiphytes in humid habitats (Barthlott et al., 2001), or on inselbergs in semi-arid climates, as observed in this study. There seems to be no correlation between high chromosome numbers in this group and adaptations to inselbergs or other habitats with extreme climates.

Within the family Commelinaceae, the genus *Tradescantia* has a basic number $x = 6$, with a predominance of diploid species, but it also exhibits intraspecific polyploidy, as in *Tradescantia blossfeldiana*, where $2n = 70, 90,$ and 110 were observed (Jones and Jopling, 1972). However, there does not seem to be any correlation between polyploidy and rupicolous habitats in *Tradescantia*, and even populations of *Tradescantia ambigua* from non-rupicolous populations had $2n = 24$ (Jones and Kenton, 1984; Pitrez et al., 2001).

The family Apocynaceae (including the Asclepiadaceae *sensu* APG III, 2009) is a karyologically stable group, with $2n = 22$ in a large majority of the species or polyploids with $2n = 44$ and 66 (Van der Laan and Arends, 1985). The present count of $2n = 20$ for *Maxillaria tenuifolia* is the first for the genus. This species is exclusive to inselbergs and rocky field vegetation in northeastern Brazil (Watanabe et al., 2009). Chromosomal counts of $2n = 20$ have been reported for other genera of Apocynaceae, such as *Strophanthus* and *Trachelospermum*, as well as $2n = 18$ for *Allamanda*, *Pachypodium*, and *Prestonia* (Van der Laan and Arends, 1985). Counts of $2n = 20$ and 22 in the 2 species examined here is within the modal range (or near the mode number) for the family, and apparently they are diploids.

The karyotypes for the 3 species of the genus *Ipomoea* (Convolvulaceae) that were analyzed here are in agreement with previous reports ($2n = 30$), including the occurrence of secondary constrictions in other species of the genus (Sinha and Sharma, 1992). Within the genus *Evolvulus*, *Evolvulus filipis* and *Evolvulus glomeratus*, both with $2n = 26$, were previously analyzed by Pitrez et al. (2008), and this chromosome number was reported for other species of this genus (Goldblatt and Johnson, 1990). The chromosome numbers observed in *Merremia aegyptia* ($2n = 30$) confirmed previously published counts (Vij et al., 1974). However,

the observations of Elias (1967), who reported $2n = 28$ for *Merremia aegyptia* and *Merremia umbellata*, were not confirmed by our observations. A more detailed discussion of the taxonomic implications and the karyotype evolution of the family Convolvulaceae were published by Pitrez et al. (2008). Overall, the chromosome numbers that were observed for species of Convolvulaceae growing on inselbergs in Paraíba do not significantly differ from those cited in the literature or from those attributed to other terrestrial species of the family. In general, the genera are karyologically stable, and their composite species do not demonstrate large variations in terms of their intra- or interspecific chromosome numbers.

Euphorbia and *Phyllanthus* (Euphorbiaceae) demonstrated wide numerical variations. Analyses of 18 species of *Phyllanthus* growing in the United States and in Central America yielded chromosome numbers that varied between $n = 8$ and $n = 78$ (Webster and Ellis, 1962), without any obvious correlation with habitat. Polyploid variation in *Euphorbia* (ranging from $2n = 18$ to $2n = 44$) was reduced among species growing on the Macaronesian Islands (Molero et al., 2002). In this case, the polyploid species were in the plesiomorphic state, while the diploids (with cactus-like growth habits adapted to environments under water stress) were the most derived. The basic numbers $x = 13$ for *Phyllanthus* and $x = 10$ for *Euphorbia* were confirmed in our survey. *Stillingia* is the least studied of the 3 genera, with only 3 published counts of $2n = 22$ (in 2 reports) and $2n = 36$ (in another) (Goldblatt and Johnson, 1990), partially coinciding with our data.

For the family Gesneriaceae, no previously published data is known for *Paliavana*, but a previous report of $2n = 26$ was found for *Sinningia incarnata* (Goldblatt, 1984). Approximately 560 of the 2500 known species of the family Gesneriaceae are epiphytes (Benzing, 1990). In Brazil, the genera *Sinningia* and *Paliavana* are predominantly rupicolous (Souza and Lorenzi, 2005), without any apparent correlation between polyploidy and adaptations to a saxicolous habitat.

Cissus sulcicaulis (Vitaceae) is frequently encountered on rock outcrops in northeastern Brazil and in terrestrial habitats in the Caatinga drylands. Although the genus *Cissus* demonstrates numerous chromosomal variations from $2n = 22$ to 96 (Moore, 1973; Goldblatt, 1984), the $2n = 34$ number cited here is unique for the genus.

Of a total of 39 angiosperm species that occur on 5 inselbergs in Paraíba State, a majority were diploids, not differing from previous chromosome records for the same species that occur in other types of habitats. Even paleopolyploid groups, such as the families Orchidaceae and Bromeliaceae demonstrated predominantly diploid cytotypes (Felix and Guerra, 2005). Groups with high levels of ploidy, such as the genera *Melocactus* (Cactaceae), *Clusia* (Clusiaceae), and *Epidendrum* (Orchidaceae), seem to be individual cases and are independent of the environmental stress assumed to be associated with inselberg habitats. In fact, for plants in areas where nutrient levels are limited, there will be selection against polyploids or for those that cannot eliminate excess DNA (Leitch and Leitch, 2008), probably because polyploidy increases demands for the production of more proteins and nucleic acids. The occurrence of these polyploid species in the inselbergs studied may be related to the efficient ability to use water. As such, this study does not support the hypothesis that polyploid species tend to occur in rupicolous habitats and, by extension, other habitats having rigorous environments.

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