

New chromosome counts in Neotropical *Machaerium* Pers. species (Fabaceae) and their taxonomic significance

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Abstract - *Machaerium* comprises about 130 species and is widespread in the Neotropics, ranging from Mexico to Argentina. Because of the existing taxonomic complexes, circumscribing many of its species is far from easy. Cytological studies on *Machaerium* have only been carried out for four species and are still meager. This paper brings forward chromosome numbers for 18 species, 16 of which represent new counts. Chromosomes were very small in size. Meiotic behavior was observed only for *M. opacum* ($n=10$) and proved to be normal. The other species showed $2n=20$, except for the tetraploids *M. nyctitans*, *M. hirtum* and *M. sericiflorum*, where $2n=40$. Polyploidy possibly evolved independently in *Machaerium* since these species belong to different sections. Both *M. hirtum* and *M. sericiflorum* (tetraploids) were regarded as *taxa* distinct from *M. aculeatum* (diploid), although they used to be considered as synonyms. The importance of polyploidy for the evolution of species in this genus and its frequency within the different sections will be assessed as more records become available. It might also be possible to confirm whether its basic number is actually $x=10$, as results tend to indicate.

Key words: chromosome numbers, cytotaxonomy, Fabaceae, *Machaerium*, polyploidy.

INTRODUCTION

Widespread in the Neotropics, the genus *Machaerium* comprises around 130 species, ranging from Mexico to Argentina, many of which are difficult to delimit taxonomically (RUDD 1986). Most diversity occurs in southeastern Brazil and in the Amazon basin. Traditionally, BENTHAM (1860) divided this genus into five series, later raised to sections by TAUBERT (1891). Closely related to *Dalbergia*, it was assigned, together with 12 other genera, to the *Dalbergia* group in the tribe Dalbergieae, subfamily Faboideae (POLHILL 1994). However, recent cladistic analyses considered this tribe was not monophyletic and distinguished a *Dalbergia* clade that only included

Machaerium and *Dalbergia*, out of the former tribe Dalbergieae, together with 14 genera from the tribe Aeschynomeneae. They were then considered as more closely related to the *Aeschynomene* species that have basifixed stipules (LAVIN *et al.* 2001).

This *Dalbergia* clade, together with other dalbergioid legumes and a group of Faboideae that also includes the tribes Amorpheae and Dipterygeae are fairly uniform cytologically and have a base chromosome number $x=10$ (GOLDBLATT 1981; LAVIN *et al.* 2001).

The only chromosome numbers reported for *Machaerium* concern *M. aculeatum* Raddi ($n=10$, COLEMAN and DE MENEZES 1980), *M. lunatum* (L.f.) Ducke ($n=9$, GILL and HUSAINI 1985a; HUSAINI and GILL 1986), from sect. Lineata; *M. acutifolium* Vogel ($2n=20$, BANDEL 1974), from sect. Acutifolia; and *M. paraguariense* ($2n=20$, SAR-

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TORI and FORNI-MARTINS, pers. com.), from sect. *Reticulata*. Therefore, information is limited to a small fraction of this genus, still poorly known, cytologically speaking.

This study aims at extending the investigation of chromosome numbers for this widespread Neotropical genus to other species and sections, trying to help circumscribing the *taxa*.

MATERIALS AND METHODS

Seeds of 17 *Machaerium* species and flower buds of *M. opacum*, presenting different life forms and from various habitats were collected in southern and north-eastern Brazil. Most of them come from areas either of forest or cerrado (a kind of savanna) (Table 1). Vouchers were housed at the Herbarium (UEC) of the Botany Department of the University of Campinas-SP. As for meiotic studies, flower buds were fixed in 3:1 ethanol:acetic acid (Carnoy) and their anthers squashed in acetocarmine (MEDINA and CONAGIN 1964). Young,

actively growing root-tips of each of the 17 species were excised from potted plants and pretreated with saturated solution of PDB (paradichlorobenzene) for 4 hours at 16-18°C, in order to obtain a better chromosomal condensation and arrangement on the slide. The material was fixed in Carnoy's liquid for 24 h, and stored in 70% ethanol. They were stained according to the HCl/Giemsa method (GUERRA 1983). An Olympus photomicroscope was used to photograph suitable chromosome spreads. Permanent slides are kept at the Biosystematic Laboratory of the Botany Department.

RESULTS

The 18 species of *Machaerium* studied belong to four out of the five sections known for the genus: sect. *Reticulata* (six species), sect. *Acutifolia* (six species), sect. *Oblonga* (three species), and sect. *Lineata* (three species). The chromosomes were very small in size making it difficult to determine karyotypes (Fig. 1, Table 1).

The chromosome analysis showed that all the species studied presented $2n=20$, except for the tetraploids *M. nyctitans*, *M. hirtum* and *M. sericiflorum*, with $2n=40$ (Fig. 1, Table 1). The last two species belong to sect. *Lineata*, and the first one to sect. *Oblonga*. Meiotic behavior proved to be normal in *M. opacum* and regular 10 bivalents were observed at diakinesis (Table 1).

DISCUSSION

The chromosome counts found represent 16 new records for *Machaerium*. The numbers for *M. acutifolium* Vogel and *M. aculeatum* Raddi ($2n=20$) confirmed those found by BANDEL (1974) and COLEMAN and DE MENEZES (1980), respectively. This number was also detected for *M. paraguariense* Vogel (SARTORI and FORNI-MARTINS, pers. com.). Exception was *M. lunatum* (L.f.) Ducke, with $n=9$ (GILL and HUSAINI 1985a; HUSAINI and GILL 1986). The basic chromosome number $x=10$ seems to fit *Machaerium* well and agrees with records about both the tribe Dalbergieae and the dalbergioid legumes (GOLDBLATT 1981; LAVIN *et al.* 2001).

Polyploidy and the taxonomy of Machaerium

The polyploidy detected for *M. nyctitans*, *M. hirtum* and *M. sericiflorum* is the first report regarding *Machaerium*. As these species belong to only two of the genus sections, sect. *Oblonga*

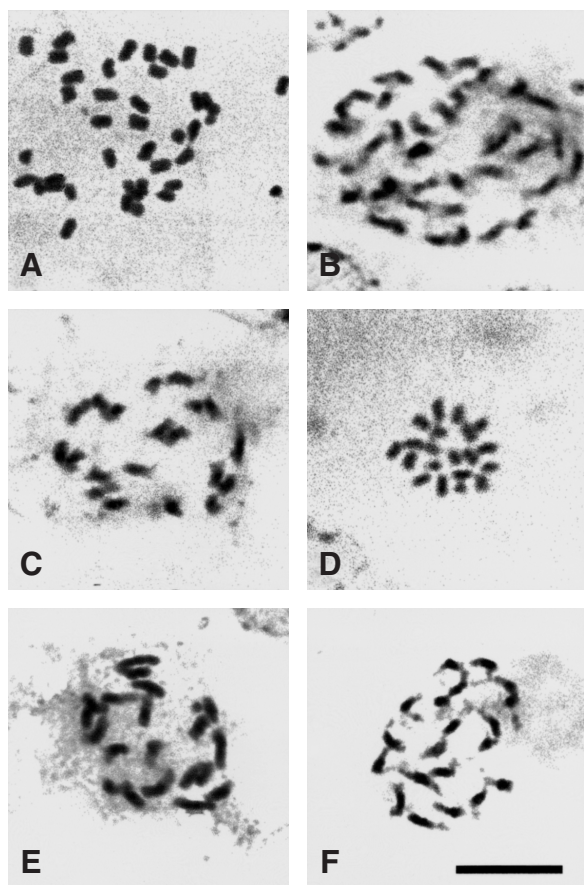


Fig. 1 – Chromosomes in *Machaerium*. A-B: tetraploid species; C-F: diploid species. A) *M. hirtum*, B) *M. sericiflorum*, C) *M. aculeatum*, D) *M. scleroxylon*, E) *M. villosum*, F) *M. punctatum*. Scale bar 10 μ m.

Table 1- Chromosome numbers and habits of the *Machaerium* species. The species were grouped into sections following BENTHAM (1860) except for those followed by (!), which have been rearranged according to MENDONÇA FILHO (in preparation).

Section/Species	habit	Voucher number (UEC)	$2n/n^*$	Ploidy level
sect. Reticulata				
<i>M. brasiliense</i> Vogel	Tree	C.V. Mendonça 443	20	2x
<i>M. lanceolatum</i> (Vogel) J.F.Macbr. (!)	Liana	C.V. Mendonça 439	20	2x
<i>M. mucronulatum</i> Mart. ex Benth. (!)	Shrub	C.V. Mendonça 658	20	2x
<i>M. oblongifolium</i> Vogel	Liana	T.S. Spósito s.n.	20	2x
<i>M. opacum</i> Vogel	Tree	C.V. Mendonça 533	10*	2x
<i>M. triste</i> Vogel	Liana	C.V. Mendonça 547	20	2x
sect. Acutifolia				
<i>M. acutifolium</i> Vogel	Tree	C.V. Mendonça 446	20	2x
<i>M. fulvovenosum</i> H.C. Lima (!)	Tree	C.V. Mendonça 600	20	2x
<i>M. pedicelatum</i> Vogel (!)	Tree	E.T. Neto s.n.	20	2x
<i>M. punctatum</i> Pers. (!)	Tree	C.V. Mendonça 548	20	2x
<i>M. stipitatum</i> Vogel	Tree	C.V. Mendonça 450	20	2x
<i>M. villosum</i> Vogel	Tree	C.V. Mendonça 434	20	2x
sect. Oblonga				
<i>M. nyctitans</i> (Vell.) Mart. ex Benth.	Tree	C.V. Mendonça 428	40	4x
<i>M. scleroxylon</i> Tul.	Tree	C.V. Mendonça 549	20	2x
<i>M. uncinatum</i> Benth. (!)	Liana	C.V. Mendonça 441	20	2x
sect. Lineata				
<i>M. aculeatum</i> Raddi	Liana	C.V. Mendonça 606	20	2x
<i>M. hirtum</i> (Vell.) Stellfeld	Tree	C.V. Mendonça 437	40	4x
<i>M. sericiflorum</i> Vogel (!)	Liana	C.V. Mendonça 633	40	4x

(*M. nyctitans*) and sect. Lineata (for the last two species), it seems reasonable to suppose that polyploidy has evolved independently within the *Machaerium* sections and may furthermore be restricted to these two sections of the genus. As for *Adesmia* (Fabaceae, Faboideae, Adesmieae), polyploidy could not be related to the serial infrageneric positions of species, which also indicates its independent origin (MIOTTO and FORNI-MARTINS 1994).

The assumption that polyploids are generally widely distributed (GUERRA 1988) was supported by the data on the first two species (very common species) but not by those on *M. sericiflorum*, which presents a more restricted distribution.

Polyploidy played an important role in the speciation of higher plants and all species of Fabaceae cytologically known to date had a polyploid ancestor (GOLDBLATT 1981; GILL and HUSAINI 1985b). However, polyploidy seems to play a secondary role in the speciation of *Machaerium* since it has only been recorded for three species, so far.

The tetraploid number found for *M. sericiflorum* and *M. hirtum* definitely separated them from *M. aculeatum* ($2n=20$). These species were classified as *M. aculeatum* by HOEHNE (1941) and many authors have followed his assumption, even recently (LORENZI 1992). Although the floral morphology of these species is similar, their fruit, seed and seedling present markedly different

characters (MENDONÇA FILHO, in preparation). Therefore, both karyotypic and morphological characters give good evidence to consider these species as distinct *taxa*.

A relationship between the species habits and their classification into sections only seems possible for *Machaerium* sect. Acutifolia, where all species are trees. As for the other sections, the habit of the species ranges from lianas, shrubs up to trees (Table 1). For the time being, no chromosomal characters could be related to the differences of habit observed within these sections. Although it has been assumed that lianas derived from trees (RADDIFORD *et al.* 1974), and shrubby plants occupy an intermediary position, trees being considered as less derived (CARLQUIST 1991), the constancy in chromosome numbers (except for the polyploids) and chromosome size observed for the *Machaerium* species studied makes it impossible to set up any cytotaxonomic considerations. LOMBELLO and FORNI-MARTINS (1998) suggested that, as for Sapindaceae, the derivation of lianas was followed by a chromosomal derivation, through enlargement of the chromosomes and reduction of their number. In addition, with regard to Malpighiaceae, another family with species showing distinct habits, the tree species *Lophanthera lactescens* showed the lowest chromosome number ($2n=12$) and the longest chromosomes (11.3 to 5.5 μm) (LOMBELLO and FORNI-MARTINS 1998).

Chromosomal data only exist for a small fraction (14%) of the about 130 species of *Machaerium*. This study must be extended to other species in order to verify the existence of a possible relationship between the derivation of karyotypic characters and changes of morphological characters.

The cytological knowledge must be used in conjunction with other sources of data, from as many different areas as possible, so as to achieve a better understanding of the phylogenetic relationship of species in the genus and come the best classification.

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