



NOTES

Chromosome number and karyotype of the endangered Amazonian woody *Centrolobium paraense* Tul. species

Nair Dahmer¹, Maria Teresa Schifino Wittmann^{1*}, and Paulo Emilio Kaminski²

Received 05 June 2006

Accepted 20 September 2009

ABSTRACT - *Centrolobium paraense* Tul., popularly known in Brazil as “pau-rainha”, is a species with a high timber potential, presently endangered due to deforestation of the Amazonian region and indiscriminate wood extraction. Chromosome number and karyotype morphology of this species are presented for the first time. All the individuals of the three populations analyzed are diploid, with $2n=2x=20$ chromosomes. The chromosomes ranging from ca. 1.7 to 4 μm in size. The karyotype is composed of three metacentric, three submetacentric (one with a satellite on the short arm), three acrocentric and one subacrocentric chromosome pairs. Other *Centrolobium* species and populations should be analyzed in order to assess the extent of intraspecific and interspecific variation in chromosome number and morphology, if any.

Key words: *Centrolobium*, “pau-rainha”, chromosome number, karyotype, Amazonian.

INTRODUCTION

The genus *Centrolobium* Mart. ex Benth. (Leguminosae, Papilionoideae), tribe Dalbergieae, is a small American tree genus, with six to seven species, naturally distributed from Panama to Ecuador, Colombia, Venezuela, Guyana, Bolivia and North and Southeastern Brazil. The main characteristic of the genus are the winged and spiny fruits, therefore the scientific name (from the greek *centron*, prickle and *lobion*, lobe, fruit) and the common name (porcupine wood, referring to the spiny fruits) (Mabberley 1997, Lewis et al. 2005) that are given to some of the species. DNA sequences positioned *Centrolobium* in the *Petrocarpus* clade (Lavin et al. 2001). The species are used mostly as timber for furniture, boat-building and carpentry in general. Among the species occurring in Brazil are *C. tomentosum*

Guill ex Benth, *C. robustum* (Vell.) Mart (popularly known as “araribá” or “araribá-rosa”) and *C. microchaete* (Mart. ex Benth.) Lima (“petimujú”) (Lorenzi 1992, 1998). Another species, with a high timber potential, is *C. paraense* Tul., popularly known as “pau-rainha”. The species occurs naturally from Panama to North Brazil, restricted to the Roraima and Pará States, and, besides wood for construction, it is reported to be used by indigenous populations as fuel, for medicinal purpose and to extract a red dye that exudes from the bark when it is cut. The trees are normally 5 to 15 m high and 50 cm BHD (breast height diameter) but may reach 30 m high and 120 BHD (Kaminski 2004). With the increasing destruction of the Amazonian tropical forest by deforestation and indiscriminate wood extraction, this species is endangered, a situation that may be

¹ Universidade Federal do Rio Grande do Sul (UFRGS), Departamento de Plantas Forrageiras e Agrometeorologia, Avenida Bento Gonçalves 7712, C.P. 15100, 91540-000, Porto Alegre, RS, Brazil. *E-mail: mtschif@ufrgs.br

² Empresa Brasileira de Pesquisa Agropecuária, Centro de Pesquisas Agroflorestais de Roraima. BR 174, Km 8, C.P. 133, 69301-970, Boa Vista, RR, Brazil

reversible with a rational management and conservation policy (Kaminski 2004).

Papers published on *Centrolobium* are mostly related to, for example, botany, ecology (Lewis and Owen 1989, Aidar and Joly 2003), management (Arco-Verde and Schwenberger 2000), analysis of biochemical compounds (Araujo et al. 1998) and inoculation (Marques et al. 2001).

To our knowledge, there is no information published on chromosome number of any *Centrolobium* species (as in Fedorov 1969, IPCN). So, this study aimed to determine the chromosome number and to construct a karyotype of *C. paraense* based on chromosome number and morphology.

MATERIAL AND METHODS

Three natural populations of the tree species *Centrolobium paraense*, managed by EMBRAPA-Roraima were analyzed: Campos Novos I, São Francisco I and São Francisco III (Table 1). The first population occurs in a transitional forest area and the other two in forest patches near cultivated areas. Fruits from each population were collected and numbered. As each fruit has from two to three seeds, one seed per fruit was used for the cytological study and the other one (or two) kept for further studies. Each seed was considered as an individual.

Somatic chromosome numbers were determined in the root-tip cells, following the technique normally used at our laboratory for other species (Reis et al. 2008,

Guisso-Navarini et al. 2008) with some adjustments. After manual scarification by removing a small fragment of the testa, the seeds were germinated in Petri dishes lined with moist filter paper at room temperature. Germination started approximately after eight days and when the roots were 0.8 to 1.0 cm long, they were pre-treated in a saturated solution of paradichlorobenzene at 4 °C for 32 hrs, fixed in 3:1 ethanol-acetic for 24 h, and stored in 70% ethanol below 0 °C until required. The root-tips were stained by the Feulgen method following a hydrolysis with 1N HCl at 60 °C for 12 min, the color was developed in Schiff reactive for 30 min, and the root-tips were then squashed in 2% propionic carmine. At least ten intact cells with well-spread chromosomes were examined per slide (one slide per seed). Results were recorded by photomicrographs and digital images.

For karyotype analysis, five cells with equivalent degrees of chromosome contraction were used. Photographs of the chromosome spreads were enlarged and the homologous chromosomes were paired by their similarity in size and shape. Short arm, long arm and total chromosome size and centromeric index (ci) (short arm length/total chromosome length) were determined and the TCL (total haploid chromosome complement length) was calculated. Chromosomes were classified as metacentric (m) (ci higher than 0.40), submetacentric (sm) (ci between 0.36 and 0.40), acrocentric (a) (ci between 0.25 and 0.35) and subacrocentric (sa) (ci lower than 0.25). The results were represented in an ideogram. Karyotype symmetry degree was calculated following Stebbins (1971).

Table 1. Chromosome number in three populations of *Centrolobium paraense*

Population /locality ^a	Individual	Number of cells analyzed	Chromosome number (2n)
Campos Novos I, Mucajaí, RR	01	10	20
	05	12	20
	10	14	20
São Francisco I, Bonfim, RR	01	12	20
	02	10	20
	18	15	20
São Francisco III, Bonfim, RR	03	11	20
	05	11	20
	06	10	20
	13	13	20
	17	25	20

^aRR - Roraima State

RESULTS AND DISCUSSION

These are the first cytogenetical information on *C. paraense* and any *Centrolobium* species. All the individuals and populations analyzed have $2n=20$ chromosomes and a similar karyotype (Table 1). The chromosomes range from ca. 1.7 to 4 μm in size and are metacentric, submetacentric, acrocentric and subacrocentric in shape (Figure 1 A and B, Table 2).

The second pair of chromosomes has a secondary constriction in the short arm, with a small satellite (sat). The karyotypic formula is $3m + 2sm + 1sm^{\text{sat}} + 3a + 1sa$. Regarding karyotype asymmetry, the *C. paraense* karyotype is classified in Stebbins (1971) category 2B (ratio of larger to smaller chromosomes between 2:1 and 4:1, and proportion of chromosomes with arm ratio smaller than 2:1 between 0.01 and 0.50) (Figure 1 A and B, Table 2).

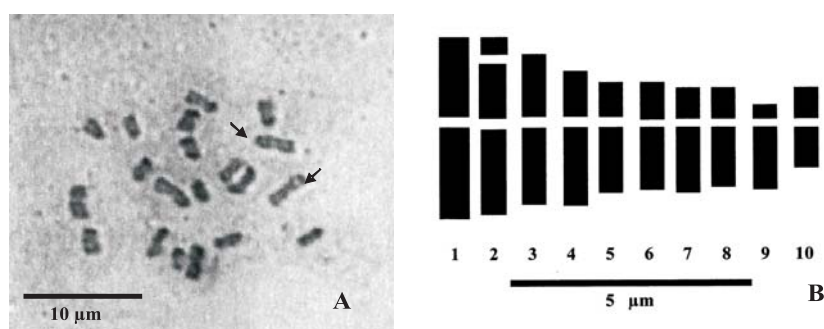


Figure 1. Somatic metaphase plate of *Centrolobium paraense* (São Francisco I population) with $2n=20$ chromosomes. Arrows point to the pair of chromosomes with secondary constrictions (A); Idiogram representing the species karyotype (B). Scale bar equal to 10 μm for A and 5 μm for B

Table 2. Karyotype data for *C. paraense*: chromosome length and centromeric index for each chromosome pair and TCL (total complement length)

Chromosome pair	1	2	3	4	5	6	7	8	9	10
Length (μm)	4.0	3.8	3.4	3.0	2.5	2.4	2.3	2.2	1.9	1.7
Centromeric index	0.45	0.38	0.44	0.39	0.32	0.39	0.33	0.35	0.18	0.43
TCL (μm)	27.20									

According to Goldblatt (1981) basic chromosome numbers in Dalbergieae are $x=10$ and $x=11$. Therefore *Centrolobium paraense* is a diploid species ($2n=2x=20$). This same number of $2n=20$ is also found in other woody genera of the Pterocarpus clade such as *Platymiscium* Vogel, *Tipuana* (Benth.) Benth. and *Geoffroea* Jacq., whereas *Pterocarpus* Jacq. has $2n=22$ (IPCN).

As a follow up to the present work, other *Centrolobium* species and populations should be analyzed in order to assess the extent of intraspecific and interspecific variation in chromosome number and morphology, if any. Considering that the genus is a small one, and three other species are known to grow naturally in Brazil, this task could be relatively easily accomplished, the only condition being the availability of material (seeds) for study.

Número cromossômico e cariótipo da espécie arbórea Amazônica *Centrolobium paraense* Tul. ameaçada de extinção *Centrolobium paraense* Tul.

RESUMO - *Centrolobium paraense* Tul., popularmente conhecida no Brasil como “pau-rainha”, é uma espécie com alto potencial madeireiro, mas ameaçada de extinção devido ao desmatamento da Amazônia e extração indiscriminada de madeira. O número cromossômico e a morfologia do cariótipo são apresentados pela primeira vez para esta espécie. Todos os indivíduos das três populações examinadas são diplóides com $2n=2x=20$ cromossomos. Os cromossomos variam de ca. 1.7

Chromosome number and karyotype of the endangered Amazonian woody *Centrolobium paraense* Tul. species

a 4 µm de tamanho. O cariótipo é composto de três pares metacêntricos, três submetacêntricos (um com um satellite no braço curto), três acrocêntricos e um subacrocêntrico. Outras espécies e populações de *Centrolobium* deveriam ser analisadas a fim de verificar a possível existência de variabilidade intraespecífica e interespecífica no número e morfologia dos cromossomos.

Palavras-chave: *Centrolobium*, pau-rainha, número cromossômico, cariótipo, Amazônia

REFERENCES

- Aidar MPM and Joly CA (2003) Dinâmica da produção e decomposição da serapilheira do araribá (*Centrolobium tomentosum* Guill. ex Benth. – Fabaceae) em uma mata ciliar, Rio Jacaré-Pepira, São Paulo. **Revista Brasileira de Botânica** **26**: 193-202.
- Araujo CAC, Alegrio LV and Leon LL (1998) Antileishmanial activity of compounds extracted and characterized from *Centrolobium sclerophyllum*. **Phytochemistry** **49**: 751-754.
- Arco-Verde MF and Schwenberger D (2000) **Plantio de espécies florestais de rápido crescimento no estado de Roraima**. Embrapa Roraima, Boa Vista, 23p. (Documentos 2).
- Federov An A (1969) **Chromosome numbers of flowering plants**. Academy of Sciences of the USSR, Leningrad, 926p.
- Goldblatt P (1981) Cytology and the phylogeny of Leguminosae. In: Polhill RM and Raven PM (eds.) **Advances in Legume Systematics 2**. Royal Botanic Gardens, Kew, p. 427-463.
- Guisso-Navarini AP, Schifino-Wittmann MT, Barros IBI and Almeida D (2008) Cytogenetics of *Hypericum caprifoliatum* Cham. & Schltdl. (Clusiaceae) populations and other species of the genus. **Crop Breeding and Applied Biotechnology** **8**: 337-345.
- IPCN (2009) Index to Plant Chromosome Numbers. Available at: <http://mobot.mobot.org/W3T/Search/ipcn.html>. Accessed on May 2009.
- Kaminski PE (2004) **O pau rainha (*Centrolobium paraense*): características, potencialidades e usos**. Embrapa Roraima, Boa Vista, 31p. (Documentos 10)
- Lavin M, Pennington RT, Klitgaard BB, Sprent JI, Lima HC and Gasson PE (2001) The dalbergioid legumes (Fabaceae): delimitation of a pantropical monophyletic clade. **American Journal of Botany** **88**: 503-533.
- Lewis GP and Owen PE (1989) **Legumes of the Ilha de Maracá**. Royal Botanic Gardens, Kew, 99p.
- Lewis G, Schrire B, Mackinder B and Lock M (2005) **Legumes of the world**. Royal Botanic Gardens, Kew, 577p.
- Lorenzi H (1992) **Árvores Brasileiras: manual de identificação e cultivo de plantas arbóreas nativas do Brasil**. Editora Plantarum, Nova Odessa, 368p.
- Lorenzi H (1998) **Árvores Brasileiras: manual de identificação e cultivo de plantas arbóreas nativas do Brasil**. Editora Plantarum, Nova Odessa, 368p.
- Mabberley DJ (1997) **The plant book**. Cambridge University Press, Cambridge, 858p.
- Marques MS, Pagano M and Scotti MRMML (2001) Dual inoculation of a woody legume (*Centrolobium tomentosum*) with rhizobia and mycorrhizal fungi in south-eastern Brazil. **Agroforestry Systems** **52**: 107-117.
- Reis CAO, Schifino-Wittmann MT and Dall'Agnol M (2008) Cytogenetic characterization of a collection of *Paspalum nicorae* Parodi accessions. **Crop Breeding and Applied Biotechnology** **8**: 212-218.
- Stebbins GL (1971) **Chromosomal evolution in higher plants**. Addison Wesley, Menlo Park, 216p.