Further Cytogenetic Information on Arachis stenosperma (Leguminosae)

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Summary Arachis stenosperma is a wild relative of the groundnut, with a peculiar disjunct distribution in Central Brazil and along the Atlantic coast. Arachis species show one pair of satellited chromosomes (SAT) and some species have a distinct, smaller pair (A pair). The SAT chromosomes are classified according to 10 types. Previous literature information on A. stenosperma documents the presence of SAT type 3 only in the coastal populations, while type 5 was restricted to accessions from Central Brazil. This work sampled a broader area of geographic distribution, represented by an increased number of accessions, and provides further information on karyotypic characteristics. All 23 accessions analyzed present 2n=20. SAT type 5 was found in 10 accessions, irrespective to their area of occurrence. So, type 5 occurs also in the Atlantic coast. Type 3, so far exclusive to the Litoral, is not the most frequent type in the area. With the present data, the potential cytogenetic distinction between coastal and continental populations has vanished. The formula 18m+2sm clearly predominates, both in the Litoral and in Central Brazilian populations. Also irrespective to the area of origin, both idiogram positions A9/SAT10 and SAT9/A10 were identified. As the SAT of type 3 is considered more primitive than type 5, and the coastal area of distribution of A. stenosperma is secondary, it's future detection in continental populations is expected. In the meantime, a unique cytogenetic feature, the presence of SAT type 3 persists as an exclusive attribute of the populations of the secondary area of occurrence.

Key words *Arachis*, Wild species, Brazil, Geographic disjunction, Karyotypes, Sattelited chromosome.

Arachis stenosperma Krapov. and W. C. Greg. is an annual, diploid, exclusively Brazilian wild relative of the groundnut, *A. hypogaea* L., associated to the A genome of this allotetraploid crop. It shows a peculiar disjunct distribution in the Brazilian Central Plateau and along the Atlantic coast (Fig. 1), obviously tied to anthropogenic migration (Simpson *et al.* 1993, Krapovickas and Gregory 1994, Valls 1996).

Present germplasm assets encompass 57 accessions of *A. stenosperma*, mostly collected, since 1968, in very disturbed sites in the Atlantic Litoral (São Paulo and Paraná states), as well as from a population found in waste places in the city of São Paulo, and from populations with variable degrees of human interference in the continental states of Mato Grosso, Goiás and Tocantins (Monçato and Valls 1999).

All *Arachis* species show at least 1 pair of satellited chromosomes (SAT), and some species of the taxonomic section *Arachis* also present a very distinct, smaller pair, usually referred to as the A pair (Husted 1933, 1936, Fernández and Krapovickas 1994). In mitotic preparations, the A pair is just slightly stained during prometaphase. Species with the A chromosome are geographically concentrated in the central area of distribution of the genus, along the borders of Central Brazil with Bolivia and Paraguay. From that area, the small chromosome is carried southwards by *A. villosa*

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Benth., and to the southwest by *A. duranensis* Krapov. and W. C. Greg. (Fernández and Krapovickas 1994). *Arachis stenosperma* carries the A pair towards the northeast and southeast, as it spreads down the valley of the Araguaia river and forms a disjunct area along the Atlantic coast, in São Paulo and Paraná States (Monçato and Valls 1999).

Based on the relative size of the satellite and the centromeric position, Fernández and Krapovickas (1994) have classified *Arachis* satellited chromosomes according to 10 types. The first karyotypic description of *A. stenosperma* based in the coastal accession HLK 410 has shown 2n=2x=20, all chromosomes being metacentric (Stalker and Dalmacio 1981). This interpretation was confirmed by Singh and Moss (1982) and Fernández and Krapovickas (1994). Although using the same accession, Cai *et al.* (1987) determined a karyotypic formula 14m+4sm+2st. In the idiogram of HLK 410, the A pair corresponds to chromosome 10 and the satellited chromosome, classified as type 3, corresponds to number 9 (Fernández and Krapovickas 1994).

Analyzing additional accessions of *A. stenoperma*, Lavia (1996, 1999) detected differences in the shape and position of chromosomes, as well as in the type of the satellited chromosome. Type 3 (called 3A in 1999) was found in the coastal populations, while type 5 occurred in accessions from Central Brazil only.

In the present work, 20 additional accessions of *A. stenosperma* have been analyzed, sampling a broader area of the geographic distribution, and providing further information on karyotypic characteristics, including the type of the satellited chromosome. Three accessions, previously studied by Lavia (1999), were also included.

Materials and methods

Seeds of *A. stenosperma* used in this work were provided by the Wild *Arachis* Genebank of Embrapa Genetic Resources and Biotechnology (Embrapa/Cenargen), located in Brasília, Distrito Federal, Brazil. Accessions used are listed in Table 1, which also gathers detailed geographic information on accessions previously studied by authors (Lavia 1996, 1999).

For mitotic preparations, root tips were excised from germinated seeds, pretreated in a solution of α -bromonaftalene for $2^{1/4}$ h at room temperature and fixed in Carnoy (ethanol: acetic acid, 3:1, v/v) for 24 h. Fixed root tips were hydrolyzed in 5 N HCl for 20 min at room temperature, and stained with Schiff solution and 2% acetocarmine. Chromosome descriptions follow the nomenclature suggested by Levan *et al.* (1964), adopting the same criteria used for *Arachis* by Fernández and Krapovickas (1994) and Lavia (1999).

Results and discussion

All accessions analyzed presented 2n=20, in agreement with previous counts for the species, and confirming data already reported for a few specific accessions (Lavia 1996, 1999). Karyotype analysis was done in 4 accessions (Fig. 2) and the type of satellited chromosome was determined in 10 accessions.

Contrary to data obtained by Fernández and Krapovickas (1994) and Lavia (1996, 1999), who reported the satellited chromosome of type 3 (or 3A) for the coastal populations, all accessions presented SAT type 5, irrespective to their area of occurrence.

In accessions Lm 1, Lm 6 and V 12575, the A chromosome corresponds to number 10, while the satellited chromosome is number 9, confirming data reported by Lavia (1999) for V 12575. In Lm 3, the A chromosome corresponds to number 9, while the satellite is located in the chromosome 10, which agrees with Lavia's (1999) description for the remaining accessions she studied.

All karyotypes show 18 metacentric and 2 submetacentric chromosomes. The submetacentric pair bears the satellite. This result agrees with Lavia's (1999) observations concerning 6 of her 7 ac-

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Table 1

Accessin	Collectors	Brazilian		Lat (S)	Long (W)	AII					Source
code (BKA)	and number	State	County/site	× ,)		karyotype formula	Type	Position	chromosome	
016071	VSSv 7382	SP	São Sebastião/Pontal da Olaria	23°46′	45°24′	10	18m+2sm		10	6	Lavia (1999)
018091	VSStGdW 7762	ΜТ	Barra do Garças/Faz. Mocambo	15°34'05"	52°12′19″	350	18m+2sm		10	9 I	Lavia (1999), present paper
020052	Jt 2	МТ	Barra do Garças/Faz. Paulista	15°32'	52°10′	340	18m + 2sm		10	6	Lavia (1999)
020176	VKSSv 9010	МТ	S. Antonio do Leverger	15°52'	56°04'	150	2n = 20				Present paper
020389	VKSSv 9017	МΤ	S. Antonio do Leverger	15°43'42"	55°41'18"	170	18m+2sm		10	6	Lavia (1999)
023001	VMiSv 10229	\mathbf{SP}	Cananéia/Bairro Carijo	25°01'21"	47°55'51"	10	18m + 2sm		10	9 I	Lavia (1999), present paper
024830	VSv 10309	МТ	Rondonópolis	16°28'	54°39′	215	18m+2sm				Present paper
030651	VGaRoSv 12488	МТ	Barra do Garça/Faz. Mocambo	15°34'31"	52°12'35"	360			10	6	Lavia (1999)
030767	VGaRoSv 12575	МТ	General Carneiro	15°42'28"	52°45'14"	360	18m + 2sm	5	6	10	Present paper
							20m	5	6	10	Lavia (1999)
030856	VSPmWiSv 13262	SP	Peruíbe/Ruina Abarebebe	24°16'25"	46°56'28"	3	2n = 20	S			Present paper
032476	VSStGdW 7805-AR	МТ	São Félix do Araguaia	11°37'51"	50°48'24"	240	2n = 20				Present paper
033367	SvSz 2411	МТ	São Félix do Araguaia	11°38'01"	50°48'39"	270	2n = 20				Present paper
033511	WPz 421	TO	Alvorada/Rio Pau Seco	12°36'50"	49°20'29"	310	2n = 20				Present paper
033529	WPz 422	TO	Araguaçu/Rio Pau Seco	12°36'58"	49°20'32"	310	2n = 20				Present paper
033596	VSPmSv 13672	МΤ	General Carneiro	15°42'18"	52°44′19″	400		5			Present paper
033936	VSPmW 13824	G	São Miguel do Araguaia/L. Alves	13°13'12"	50°34'38"	280	2n = 20	5			Present paper
033944	VSPmW 13828	GO	São Miguel do Araguaia/L. Alves	13°12'46"	50°35'07"	280	2n = 20	5			Present paper
033961	VSPmW 13832	GO	São Miguel do Araguaia/L. Alves	13°13'05"	50°34'07"	280	2n = 20				Present paper
033987	VSPmW 13844	TO	Araguaçu/Rio Pau Seco	12°36'51"	49°20'33"	310	2n = 20				Present paper
035246	SvW 3711	МТ	Cocalinho	14°41'23″	51°11'53"	290	2n = 20				Present paper
035254	SvW 3712	МΤ	Cocalinho	14°22'44″	51°00′10″	220	2n = 20				Present paper
035998	Lm 1	PR	Matinhos/Caiobá	25°50'11"	48°32'26"	5	18m + 2sm	5	6	10	Present paper
036005	Lm 2 and 3	PR	Antonina	25°27'49″	48°41'16"	5	18m + 2sm	5	10	6	Present paper
036013	Lm 5	PR	Antonina	25°25'59″	48°42'51"	10	2n = 20	5			Present paper
036625	Lm 6	PR	Paranaguá/Ilha da Cotinga	25°30'	48°29′	5	18m + 2sm	5	6	10	Present paper
036633	Lm 7	PR	Paranaguá/Ilha da Cotinga	25°30'	48°29′	5	2n = 20	5			Present paper
037516	V 14474	$^{\mathrm{SP}}$	São Paulo/Cid. Universitária	23°33'37"	46°43'43"	800	2n = 20				Present paper

cessions studied. However, Lavia (1999) found 20 metacentric chromosomes in V 12575, also studied here. New analyses of this accession, involving more individual plants, may allow for a better interpretation of this feature, or may document variability for the character in a single population.

The study of 5 accessions from Mato Grosso State and 3 from the coastal area by Lavia (1999) and Fernández and Krapovickas (1994) has established a geographic distinction concerning the satellited chromosome, type 3 (or 3A) in the coastal area, and type 5 in Central Brazilian accessions. But, the present analysis of additional accessions has confirmed the occurrence of type 5 also in the Atlantic

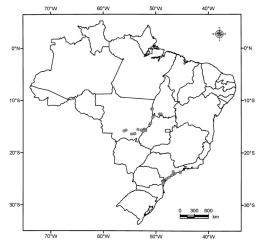
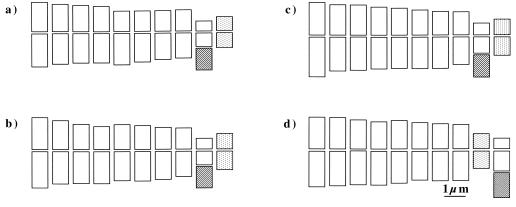


Fig. 1. Location of accessions of *Arachis stenosperma* in the Brazilian territory.



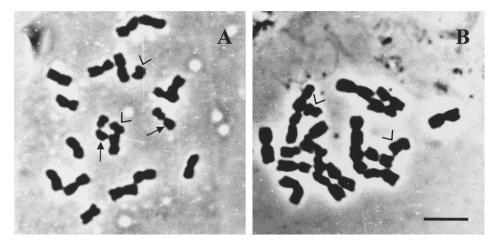


Fig. 3. Mitotic metaphases of *A. stenosperma*: a) accession and b) accession Lm 6 V 12575. Arrows indicates satellite and arrowheads indicate 'A' chromosomes.

coast (Fig. 3). Although type 3 is so far exclusive to the Litoral, it is clearly not the most frequent regional type.

With the present data, the potential cytogenetic distinction between coastal and continental populations has vanished, raising interesting implications. Single accessions with 20 metacentric chromosome have been described from both areas, although our observations on the Central Brazilian accession V 12575 are not in agreement with previous information. The formula 18m+2sm clearly predominates, both in the Litoral and in Central Brazilian populations. Also irrespective to the area of origin, both arrangements of chromosome position, A9/SAT10 (the most frequent) and SAT9/A10, were identified.

As the satellited chromosome of type 3 (or 3A) is considered more primitive than type 5, the future detection of SAT type 3 in continental populations is most probable, once the coastal area of distribution of *A. stenosperma* is considered secondary. The appearance of type 3 in the coastal populations after migration from Central Brazil would imply a regressive structural change, from an advanced to a primitive type, certainly an event not easy to explain.

On the other hand, the eventual confirmation of occurrence of Central Brazilian populations of *A. stenosperma* with the satellited chromosome of type 3 would imply parallel human transportation of the distinct cytotypes, not necessarily simultaneous, and most certainly from distinct geographic sources. It is noteworthy to add that *A. stenosperma* populations from the coastal area are considered to be more homogenous than those of the central area, on morphological and molecular grounds (Monçato 2000), as a consequence of genetic narrowing during migration in the hands of man. However, a unique cytogenetic feature, the satellited chromosome of type 3 is, so far, an exclusive attribute of the populations in the secondary area of occurrence.

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