KARYOLOGY

Karyotype analysis in several South American species of *Solanum* and *Lycianthes rantonnei* (Solanaceae)

M. Cristina Acosta[1], Gabriel Bernardello[1], Marcelo Guerra[2] & Eduardo A. Moscone[1]

[1] *Instituto Multidisciplinario de Biología Vegetal (IMBIV), CONICET-Universidad Nacional de Córdoba, Casilla de Correo 495, 5000 Córdoba, Argentina. mcacosta@imbiv.unc.edu.ar, bernarde@imbiv.unc.edu.ar, moscone@imbiv.unc.edu.ar (author for correspondence)*

[2] *Departamento de Botânica, Centro de Ciências Biológicas, Universidade Federal de Pernambuco, 50670-420 Recife, PE, Brazil. mguerra@npd.ufpe.br*

Somatic chromosomes of 17 species belonging to 4 subgenera and 7 sections of *Solanum* and *Lycianthes rantonnei* are studied. All taxa have $2n = 24$. The chromosome numbers of *S. tripartitum*, *S. tenuispinum*, and *S. vaillantii* are reported for the first time, and the karyotype analysis of *L. rantonnei* is the first one for the genus. Morphometric chromosome analyses bring estimates of karyotype composition and asymmetry. In general, karyotypes are symmetrical with the chromosomes smaller than 4 µm long, being metacentric (69%), submetacentric (24%), or more rarely subtelocentric (7%). *Solanum tucumanense*, *S. palinacanthum*, and *Solanum* sp. (*Acanthophora*) are unique by having mostly sm and st chromosomes. All species have nucleolar organizing regions and attached satellites of variable size on short arms of one chromosome pair, usually sm or st. All species can be cytologically distinguished. Karyotype data do not allow separation of *L. rantonnei* from species of *Solanum*. Results support the validity of *S. tucumanense* with respect to *S. pseudocapsicum*. Using chromosome data exclusively, cluster analysis in 10 species from *Solanum* subgen. *Leptostemonum* reinforce the systematic arrangement of taxa in subsections based on morphological features. Because of its increased karyotype asymmetry, section *Acanthophora* appears to be an advanced taxon within subgen. *Leptostemonum*.

KEYWORDS: classical chromosome staining, karyotype evolution, karyosystematics, *Lycianthes*, phenetic analysis, South American Solanaceae, *Solanum*.

INTRODUCTION

Solanum L. is one of the largest genera of angiosperms with at least 1033 known species, nearly 75% of them growing in South America (Hunziker, 2001). It includes plants of economical value, some of them cultivated for their edible tubers, fruits or leaves, such as *S. tuberosum* L. (potato), one of the most important food crops all over the world, *S. melongena* L. (eggplant), *S. muricatum* Aiton (pepino), and *S. quitoense* Lam. (lulo). Furthermore, many species have pharmaceutical interest as source of steroidal compounds, e.g., *S. viarum* Dunal and *S. elaeagnifolium* Cav., and ornamental uses, e.g., *S. pseudocapsicum* L. (Nee, 1991; Edmonds & Chweya, 1997; Hawkes, 1999). On the other hand, *Lycianthes* (Dunal) Hassler is a taxon sometimes submerged under *Solanum*, whose validity as an independent genus has been justified recently (cf. Bitter, 1919; Hunziker, 2001). It comprises around 150 species mostly neotropical, one of them, *L. rantonnei* (Carrière) Bitter, cultivated as ornamental.

Although much cytogenetic information by classical staining techniques is available in *Solanum*, still more than half of its species remains karyologically unknown (Hunziker, 2001). Most of the cytological work refers to chromosome number reports and studies on meiosis, particularly in tuber-bearing species and hybrids (cf. Howard, 1970; Edmonds, 1977; Moscone, 1989a, 1992; Hawkes, 1990; Ochoa, 1990, 1999; and papers cited herein). Comparative karyotype studies are scarce (Oinuma, 1949; Magoon & al., 1962; Mitra, 1967; Wu & Li, 1985; Okoli, 1988; Badr & al., 1997) and in general they lack qualitative or quantitative analysis, except those comprehensive contributions on sect. *Basarthrum* (Bitter) Bitter by Bernardello & Anderson (1990) and sect. *Lasiocarpa* (Dunal) D'Arcy by Bernardello & al. (1994). Concerning *Lycianthes*, only five species [*L. arrazolensis* (J. M. Coult. & Donn. Sm.) Bitter, *L. jalicensis* E. A. Dean, *L. lycioides* (L.) Hassler, *L. moziniana* (Dunal) Bitter, and *L. rantonnei*] have been cytologically explored up to now, and as yet no karyotypic study has been done in the genus (cf. Hunziker, 2001).

As in most genera of subfamily Solanoideae Schltdl., the basic chromosome number of *Solanum* and

Lycianthes is usually $x = 12$, mainly found at the diploid level ($2n = 2x = 24$), although in the first genus polyploid series are frequent ($2n = 4x = 48$, $2n = 6x = 72$, $2n = 8x = 96$). In addition, a few *Solanum* species display anomalous figures, i.e., $x = 11$ (*S. mammosum* L.), 15 (*S. bullatum* Vell.), and 23 (seven species of *Solanum* subgen. *Archaeosolanum* Marzell). The karyotypes in *Solanum* are generally composed of homomorphic chromosomes of small size (less than 4 µm), except in two sections of controversial placement in the genus, *Cyphomandropsis* Bitter and *Pachyphylla* (Dunal) Dunal, where the chromosomes have greater dimension (from 4 to 10 µm) (Roe, 1967; Stebbins, 1971; Moscone, 1989a, 1992; Pringle & Murray, 1991; Bohs, 1994, 2001; Hunziker, 2001).

In the present contribution, a morphometric karyotype analysis has been performed in 17 species of *Solanum* belonging to four subgenera and seven sections and *Lycianthes rantonnei*, all of them native to South America, as a part of a broad karyosystematic study on *Solanum* and related taxa. The aims of this paper are to: (1) report karyotype data on the largest genus *Solanum* and close allies, (2) contribute to the cytogenetic characterization of the species, (3) suggest taxonomic relationships among species, considering that the boundaries and infrageneric classification of *Solanum* have been partly conflicting, and (4) explore patterns of chromosomal differentiation and karyoevolutionary trends.

MATERIALS AND METHODS

The provenance of the plant material studied is shown in Table 1. The respective voucher specimens were identified by A. T. Hunziker, G. E. Barboza, and M. Matesevach, and deposited in the herbarium of the Botanical Museum of Cordoba, Argentina (CORD). In *Solanum*, the subgeneric and sectional system proposed

Table 1. List of species studied, code, provenance and voucher specimen.

Taxon[a]	Code	Provenance and voucher specimen[b]
Lycianthes rantonnei (Carrière) Bitter (19, 91)	Lyc	Argentina: Prov. Misiones, Dept. Candelaria, Santa Ana; *RS, EAM 4260*
Solanum L.		
Subgen. *Solanum*		
Sect. *Solanum*		
S. chenopodioides Lam. (9, 18)	che	Argentina: Prov. Córdoba, Dept. Calamuchita, Villa Quillinzo; *MCA 11*
Sect. *Pseudocapsicum* (Moench) Roem. et Schult.		
S. pseudocapsicum L. (5, 32)	pse	Argentina: Prov. Córdoba, Dept. Río Cuarto, Alpa Corral; *EAM 242*
S. tucumanense Griseb. (22, 72)	tuc	Argentina: Prov. Catamarca, Dept. Paclín, Cuesta del Totoral; *ATH, RS, LMB 24721*
Subgen. *Potatoe* (G. Don) D'Arcy		
Sect. *Etuberosum* (Bukasov & Kameraz) A. Child		
S. palustre Schltdl. (10, 26)	palu	Argentina: Prov. Neuquén, Dept. Lácar, Quila Quina; *LMB*, *EAM 573*
Sect. *Dulcamara* Dumort.		
S. endoadenium Bitter (15, 41)	end	Argentina: Prov. Tucumán, Dept. Tafí, El Molle; *ATH, GEB, EAM 24875*
S. tripartitum Dunal (21, 52)	tri	Argentina: Prov. Salta, Dept. La Poma, Cuesta de Muñano; *ATH, RS, LMB 24745*
Subgen. *Brevantherum* (Seithe) D'Arcy,		
Sect. *Holophylla* (G. Don) Walp.		
S. argentinum Bitter & Lillo (8, 36)	arg	Argentina: Prov. Córdoba, Dept. Capital, Córdoba; *MCA 20*
Subgen. *Leptostemonum* (Dunal) Bitter		
Sect. *Melongena* (Mill.) Dunal		
Subsect. *Cryptocarpum* (Dunal) G. Don		
S. sisymbriifolium Lam. (2, 9)	sis	Argentina: Prov. Córdoba, Dept. Calamuchita, Villa Quillinzo; *MCA 3*
Solanum sp. 1 (7, 30)	Sol1	Brazil: Est. Minas Gerais, between Ipanema and Manhuaçu; *ATH 25131*
Subsect. *Lathyrocarpum* G. Don		
S. elaeagnifolium Cav. (12, 67)	ela	Argentina: Prov. Córdoba, Dept. Tulumba, near Lucio V. Mansilla; *EAM, GEB 34*
S. euacanthum Phil. (20, 157)	eua	Argentina: Prov. Córdoba, Dept. Tulumba, near Lucio V. Mansilla; *LMB, LG 523*
Sect. *Acanthophora* Dunal		
Subsect. 2		
S. atropurpureum Schrank (10, 45)	atr	Argentina: Prov. Misiones, Dept. Iguazú, Puerto Bossetti; *EAM, JRD 221*
S. capsicoides All. (18, 101)	cap	Brazil: Est. Rio de Janeiro, Munic. Petropolis, Belvedere; *ATH s.n.*
S. tenuispinum Rusby (18, 80)	ten	Argentina: Prov. Tucumán, Dept. Monteros, La Heladera; *ATH, GEB, EAM 24870*
S. vaillantii Dunal (11, 56)	vai	Brazil: Est. Espiritu Santo, Munic. Conceçao de Castelho, between Castelho and Caxixe Quente; *ATH 25134 bis*
Subsect. *Acanthophora*		
S. palinacanthum Dunal (30, 150)	pali	Argentina: Prov. Catamarca, Dept. Paclín, La Merced; *ATH, RS, LMB 24713*
Solanum sp. 2[c] (7, 63)	Sol2	Argentina: Prov. Misiones, Dept. Iguazú, between Puerto Paulito and Puerto Bossetti; *RS, EAM 4160*

[a] In parentheses are the number of seedlings and somatic metaphases analysed per species, respectively.
[b] Abbreviations: Prov. = province, Dept. = department, Est. = state, Munic. = municipality; *MCA* = M. C. Acosta, *GEB* = G. E. Barboza, *LMB* = L. M. Bernardello, *JRD* = J. R. Daviña, *LG* = L. Galetto, *ATH* = A. T. Hunziker, *EAM* = E. A. Moscone, *RS* = R. Subils.
[c] Taxon without assigned subsection.

by Hunziker (2001) has been followed; as he did not include species lists, we used Morton's (1976) treatment to arrange species alphabetically into sections. For subgen. *Leptostemonum* and section *Etuberosum* the classifications of Nee (1999) and Contreras-M. & Spooner (1999) were utilized, respectively.

Primary roots obtained by germinating seeds were used to study somatic chromosomes. In some cases, 500–1000 ppm gibberellic acid (GA_3) was applied for breaking dormancy (Ellis & al., 1985). Root tips were pretreated either with paradichlorobenzene-saturated solution for 2 h at room temperature or with 2 mM 8-hydroxyquinoline for 16 h at 4ºC, and then, fixed in 3:1 ethanol:acetic acid mixture for a minimum of 12 h. Meristem cells were isolated, macerated, and squashed in a drop of 45% acetic acid after staining either with alcoholic hydrochloric acid carmine (Snow, 1963) for 3–4 days or with 2% Giemsa (Guerra, 1983) for 10 min. Slides were made permanent without removing the coverslip (Bradley, 1948) for the first staining method, or removing it by freezing with liquid nitrogen for the second one. Comparisons of chromosome size on test preparations of *S. endoadenium* and *S. elaeagnifolium* stained either with carmin or Giemsa demonstrated no differences between both staining procedures.

A total of 244 individuals from 18 samples were analyzed. From 9 to 157 cells per species were examined under a Leica DMLB microscope (cf. Table 1). Five to 11 metaphase plates from 2–8 individuals of each species were photographed with a Leica DC 250 digital camera, and the photographs used to take measurements of the following features for each chromosome pair: s (short arm length), l (long arm length), and c (total chromosome length). The arm ratio (r = l/s) was calculated and utilized to classify the chromosomes as recognized by Levan & al. (1964) as: m - metacentric (r = 1.00–1.69), sm - submetacentric (r = 1.70–2.99), st - subtelocentric (r = 3.00–6.99), and t - telocentric (r = 7.00 and up). Battaglia's (1955) terminology for satellites was used as follows: microsatellite, having less diameter than the chromosome diameter and small size; macrosatellite, having equal diameter than the chromosome diameter and large size; linear satellite, having the shape of a long chromosomal segment. The satellite lengths were added to the lengths of the corresponding arms, except in the case of the linear satellites of *S. euacanthum*. In addition, haploid karyotype length (HKL) based on the mean chromosome lengths for each species, average chromosome length, average arm ratio, and ratio between the longest and the shortest chromosome of the complement (R) were calculated. Idiograms were based on the mean values for each species. The chromosomes were arranged first into groups according to their increasing arm ratio (from m to st), and then according to the decreasing length within each group. Chromosome markers allowed sure identification of several chromosome pairs. As certain chromosomes showed great similarity, some homologies were tentatively established and others were prevented. In the last case, chromosomes were grouped in the idiograms. Karyotype asymmetry was estimated using the following parameters: the intrachromosomal asymmetry index $A_1 = 1 - [(\Sigma\, b_i/B_i) / n]$ (b_i = mean short arm length of each chromosome pair, B_i = mean long arm length of each chromosome pair, n = number of chromosome pairs), which indicates the length difference among the chromosome arms, and the interchromosomal asymmetry index A_2 = s/x (s = standard deviation, x = mean chromosome length), which indicates the size variation

Table 2. Karyotype features of *Lycianthes rantonnei* and 17 species of *Solanum*, all with 2*n* = 24[a].

Taxon	HKF	chr-NOR	HKL	c (sd)	chr-l.	r (sd)	R	A_1	A_2	AT
L. rantonnei	11 m + 1 sm	12 (sm)	26.38 (1.93)	2.20 (0.13)	1.99–2.46	1.24 (0.21)	1.24	0.18	0.06	1A
S. chenopodioides	9 m + 3 sm	10 (sm)	16.56 (0.36)	1.38 (0.06)	1.29–1.45	1.42 (0.56)	1.12	0.22	0.04	2A
S. pseudocapsicum	9 m + 2 sm + 1 st	12 (st)	20.64 (3.06)	1.72 (0.18)	1.48–2.07	1.45 (0.66)	1.40	0.22	0.11	2A
S. tucumanense	4 m + 6 sm + 2 st	12 (st)	20.42 (2.55)	1.70 (0.20)	1.37–2.05	2.14 (1.13)	1.50	0.44	0.12	2A
S. palustre	7 m + 4 sm + 1 st	8 (sm)	16.30 (0.64)	1.36 (0.19)	1.12–1.76	1.63 (0.65)	1.57	0.31	0.14	2A
S. endoadenium	8 m + 4 sm	9 (sm)	26.92 (3.08)	2.25 (0.14)	1.99–2.43	1.52 (0.45)	1.22	0.29	0.06	2A
S. tripartitum	8 m + 3 sm + 1 st	9 (sm)	19.07 (0.81)	1.59 (0.17)	1.31–1.91	1.56 (0.59)	1.46	0.30	0.11	2A
S. argentinum	8 m + 3 sm + 1 st	12 (st)	23.40 (2.94)	1.95 (0.17)	1.66–2.19	1.52 (0.61)	1.32	0.27	0.09	2A
S. sisymbriifolium	9 m + 3 sm	12 (sm)	38.75 (5.61)	3.23 (0.35)	2.71–3.76	1.42 (0.50)	1.39	0.24	0.11	2A
Solanum (*Melongena*) sp. 1	11 m + 1 sm	11 (m)	35.96 (1.84)	3.00 (0.31)	2.54–3.43	1.21 (0.19)	1.35	0.16	0.10	1A
S. elaeagnifolium	10 m + 2 sm	11 (sm)	20.83 (1.28)	1.74 (0.20)	1.42–2.09	1.32 (0.30)	1.47	0.21	0.12	2A
S. euacanthum	10 m + 2 sm	10 (m)	18.43 (0.50)	1.54 (0.18)	1.33–1.94	1.30 (0.31)	1.46	0.20	0.12	1A
S. atropurpureum	10 m + 2 sm	12 (sm)	19.12 (3.44)	1.60 (0.21)	1.36–2.08	1.31 (0.37)	1.53	0.19	0.13	2A
S. capsicoides	9 m + 3 sm	10 (sm)	23.72 (0.79)	1.98 (0.23)	1.63–2.47	1.50 (0.52)	1.52	0.28	0.12	2A
S. tenuispinum	7 m + 4 sm + 1 st	8 (sm)	20.81 (0.99)	1.73 (0.27)	1.36–2.39	1.69 (0.69)	1.76	0.33	0.16	2A
S. vaillantii	9 m + 2 sm + 1 st	12 (st)	18.09 (0.87)	1.51 (0.17)	1.28–1.90	1.66 (1.16)	1.48	0.27	0.11	2A
S. palinacanthum	4 m + 2 sm + 6 st	7 (st)	29.72 (2.71)	2.48 (0.36)	1.99–3.29	2.56 (1.19)	1.65	0.50	0.15	3A
Solanum (*Acanthophora*) sp. 2	5 m + 5 sm + 2 st	11 (st)	24.60 (3.12)	2.05 (0.56)	1.46–2.96	1.91 (0.88)	2.03	0.38	0.27	2B

[a] Species are listed following order of Table 1. Abbreviations: HKF = haploid karyotype formula; m = metacentric; sm = submetacentric; st = subtelocentric; NOR = nucleolus organizing region; chr-NOR = ordering number of the NOR-bearing chromosome and type; HKL = haploid karyotype length in µm - mean (sd); sd = standard deviation; c = mean chromosome length; chr-l = range of chromosome length; r = mean arm ratio; R = ratio between the longest and the shortest chromosome pair; A_1 = intrachromosomal asymmetry index; A_2 = interchomosomal asymmetry index; AT = Stebbins' karyotype asymmetry type.

among the chromosomes (Romero Zarco, 1986), and Stebbins' (1971) karyotype asymmetry categories.

In the species studied of *Solanum* subgen. *Leptostemonum*, a phenetic analysis was performed by using four variables per genome: HKL, R, A_1, and A_2. INFOSTAT version 1.1 (Infostat Group, 2002) was used to standardize a data matrix of the cited variables, to calculate the average Euclidean distance, to generate a UPGMA (unweighted pair-group method using arithmetic averages) phenogram and to obtain a cophenetic correlation coefficient (Sneath & Sokal, 1973).

RESULTS

The somatic chromosome number $2n = 24$ is found in all taxa examined. The chromosomes are small (Table 2, Figs. 1, 2). The average chromosome length (haploid karyotype length) is 2.20 (26.38) µm for *Lycianthes rantonnei* and 1.93 (23.14) µm for the *Solanum* species as a whole, with averages for individual species ranging from 1.36 (16.30) µm in *S. palustre* to 3.23 (38.75) µm in *S. sisymbriifolium*. The shortest measured chromosome is pair no. 12 of *S. palustre* (1.12 µm) and the longest is pair no. 1 of *S. sisymbriifolium* (3.76 µm) (Table 2). *Solanum* (*Melongena*) sp. 1 and *S. sisymbriifolium* have larger chromosomes, with length ranges of 2.54–3.43 and 2.71–3.76 µm, respectively.

All species have one chromosome pair carrying nucleolus organizing regions (NORs) plus attached satellites in the short arms. Usually the NOR-bearing pair is one of the six largest pairs of the karyotype (in 12 *Solanum* species) and most commonly the second one (6 species), but in the members of *Solanum* sect. *Pseudocapsicum* and subsect. *Cryptocarpum* (sect. *Melongena*) it is the smallest pair (Fig. 3). The NOR-bearing pair can be sm (10 species) or st (6 species), except in *S. euacanthum* and *Solanum* (*Melongena*) sp. 1 where it is m (Table 2). The frequency of appearance of the satellites varies according to the taxa. In most species, satellites are found in 85–100% of the metaphases; however, in some entities they are visible in lower percentages: *S. tucumanense* (50%), *S. elaeagnifolium* (60%), *S. tripartitum* (65%), and S *endoadenium* (70%). Usually, satellites are seen in both members of the respective chromosome pair, although in the last three cited taxa, they frequently appear in just one homologue (Figs. 1F, 2C). Satellites have constant size in *S. tucumanense* (microsatellites, Fig. 1D), *S. palustre* (macrosatellites, Fig. 1E), and the members of subsect. 2 (sect. *Acanthophora*), i.e., *S. atropurpureum*, *S. capsicoides* and *S. vaillantii* (microsatellites, Fig. 2E, F, H), and *S. tenuispinum* (macrosatellites, Fig. 2G). In the remaining species, satellite length varies and different size combinations per cell between and, often, within individuals are found, the most frequent being the heterozygous condition with one homologous chromosome bearing a microsatellite and the other one carrying a macrosatellite (Figs. 1A, 2B). In *S. euacanthum*, satellites are extremely variable in size, and appear as micro- (12%), macro- (78%), and linear satellites (12%), which could be as large as the rest of the corresponding chromosome (Fig. 2D).

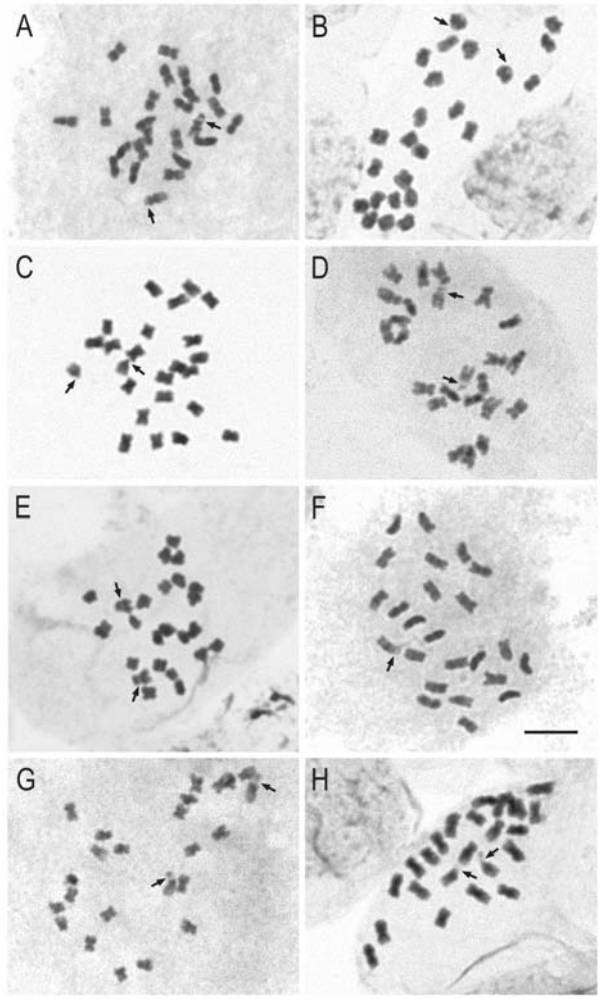

Fig. 1. Somatic metaphases of *Lycianthes* and *Solanum* species ($2n = 24$). A, *L. rantonnei*; B, *S. chenopodioides*; C, *S. pseudocapsicum*; D, *S. tucumanense*; E, *S. palustre*; F, *S. endoadenium*; G, *S. tripartitum*; H, *S. argentinum*. Arrows indicate nucleolar organizing regions. Bar = 5 µm.

Species can be distinguished by a combination of karyotype formula and position of NORs in a particular chromosome pair (Table 2, Fig. 3). Karyotype length and asymmetry indexes are also useful to discriminate some taxa. In general, karyotypes are symmetrical considering both centromere position and chromosome size variation (Table 2, Fig. 3). Most species (15) have from 7 to 11 m chromosome pairs in their diploid complements.

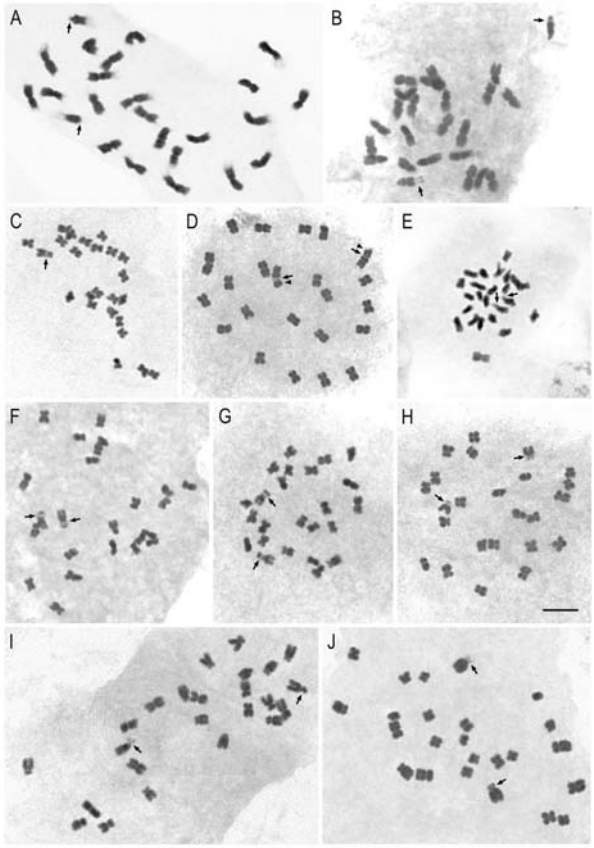

Fig. 2. Somatic metaphases of *Solanum* subgen. *Leptostemonum* species (*2n* = 24). A, *S. sisymbriifolium*; B, *Solanum* (*Melongena*) sp. 1; C, *S. elaeagnifolium*; D, *S. euacanthum*; E, *S. atropurpureum*; F, *S. capsicoides*; G, *S. tenuispinum*; H, *S. vaillantii*; I, *S. palinacanthum*; J, *Solanum* (*Acanthophora*) sp. 2. Arrows indicate nucleolar organizing regions. Arrowheads mark linear satellites. Bar = 5 µm.

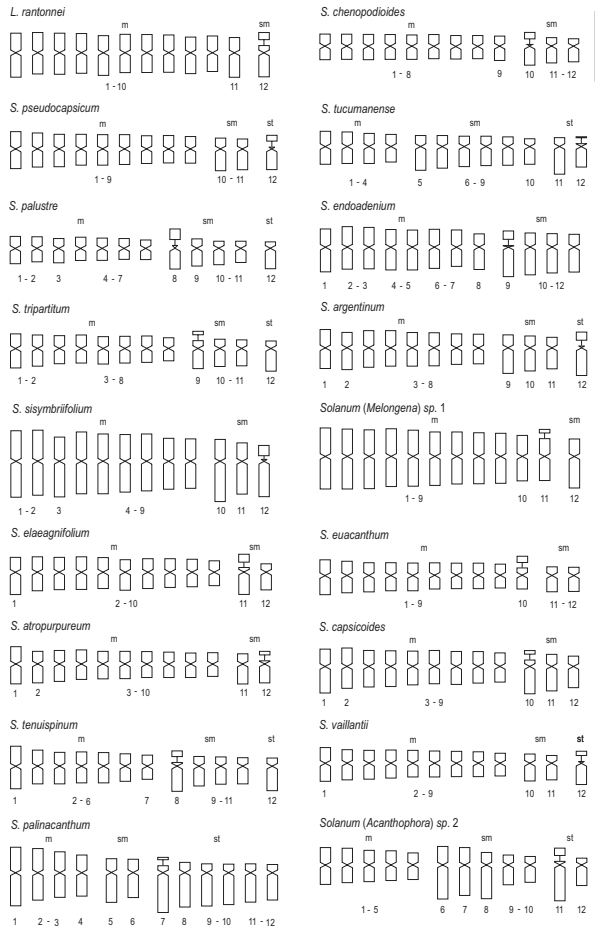

Fig. 3. Idiograms of *Lycianthes rantonnei* and *Solanum* species. Taxa ordered as in Table 1. In the different species, chromosomes that have the same number on the idiograms are not necessarily homeologous. Bar = 4 µm.

According to the mean arm ratio, they fall into the m category, although 7 of them show high r values for the category (1.50–1.69). On the other hand, *S. tucumanense*, *S. palinacanthum* and *Solanum* (*Acanthophora*) sp. 2 are distinct in possessing karyotypes mostly composed of sm and st chromosomes, and are placed into the sm category. This fact is reflected by their highest A_1 values. Overall, m chromosomes are the most common (69% of the chromosomes from all taxa), followed by sm and st chromosomes, which are less frequent (24% and 7%, respectively). Telocentric chromosomes are absent. Arm ratios found in m and sm chromosomes of the species studied cover the whole range recognized for these categories, while st chromosomes display an r range of 3.05–5.37. *Lycianthes rantonnei* and *Solanum* (*Melongena*) sp. 1 exhibit notably low variation in r estimates and, in contrast, *S. tucumanense*, *S. vaillantii*, and *S. palinacanthum* are very variable.

Asymmetry has been estimated by the A_1 and A_2 indices, which reach values for individual species in a range of 0.16–0.50 and 0.04–0.27, respectively (Table 2, Fig. 4). The karyotypes of *Solanum* (*Acanthophora*) sp. 2 ($A_1 + A_2$ values = 0.65), *S. palinacanthum* (0.65), and *S. tucumanense* (0.56) are the most asymmetrical, whereas those of *L. rantonnei* (0.24), *S. chenopodioides* (0.26), and *Solanum* (*Melongena*) sp. 1 (0.26) are the most symmetrical. In particular, *Solanum* (*Acanthophora*) sp. 2 with the highest R and A_2 index estimates, is unique in having a bimodal karyotype composed of four large (nos. 6, 7, 8, and 11) and eight small chromosome pairs. Conversely, *S. chenopodioides* with the lowest R and A_2 index values, displays all chromosomes of similar size. Following Stebbins' (1971) karyotype asymmetry classification, most species fall into category 2A by possessing mainly metacentric chromosomes of homogeneous size (Table 2). Remarkable exceptions are *S. palinacanthum*

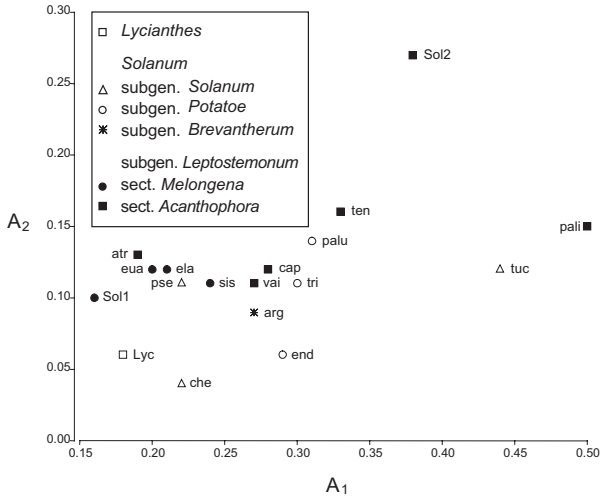

Fig. 4. Diagram showing the intrachromosomal asymmetry index (A_1) plotted against the interchromosomal asymmetry index (A_2). Species codes given in Table 1.

(3A), which has mostly st chromosomes, and *Solanum* (*Acanthophora*) sp. 2 (2B), with high chromosome size variation. No association between karyotype length and asymmetry can be established (see Table 2, Fig. 4).

As several species of *Solanum* subgen. *Leptostemonum* (10) are included in the present contribution, we have analysed the obtained data by numerical methods. The cophenetic correlation for the UPGMA phenogram is 0.934, indicating a very good fit between the cophenetic value matrix and the average Euclidean distance matrix. Dissimilarity values between species range from 0.17 to 2.13 (Fig. 5). The phenogram based on karyotype features shows the following clusters: (1) both species of subsect. *Cryptocarpum* separated with larger chromosomes than in the remainder taxa; (2) species of subsect. *Lathyrocarpum* and subsection no. 2

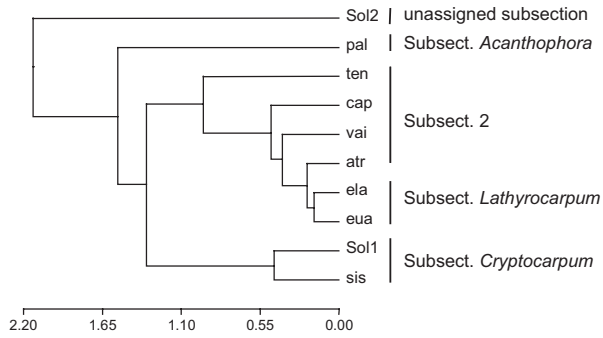

Fig. 5. UPGMA phenogram derived from average Euclidean distance between species of *Solanum* subgen. *Leptostemonum*. Species codes given in Table 1. Subsections after Nee (1999). Scale indicates dissimilarity values.

of sect. *Acanthophora*, in the larger and less homogeneous cluster, with mostly m chromosomes and rather similar chromosome length and A_2 asymmetry index estimates, both species of the first subsection exhibiting very similar karyotype features; (3) *Solanum palinacanthum*; and (4) *Solanum* (*Acanthophora*) sp. 2 both isolated with their asymmetrical karyotypes, the former possessing predominantly st chromosomes (highest A_1 asymmetry index value) and, the latter, displaying a majority of sm chromosomes and a bimodality in chromosome sizes (highest A_2 asymmetry index value).

DISCUSSION

General karyotype characteristics. — *Lycianthes rantonnei* and the *Solanum* species examined are diploids with $x = 12$, the most common basic number in the family being present in more than half of the species studied up to now (Hunziker, 2001). The chromosome numbers of *S. tripartitum*, *S. tenuispinum*, and *S. vaillantii* are reported here for the first time. In spite of the diploid ploidy level here reported, polyploidy is a relevant phenomenon in some subgenera of *Solanum*, i.e., *Solanum* (sect. *Solanum*; Edmonds, 1977), *Potatoe* (sect. *Petota* Dumort.; Hawkes, 1990; Ochoa, 1990, 1999), *Archaeosolanum* (Randell & Symon, 1976), and *Leptostemonum* (sect. *Melongena* subsect. *Lathyrocarpum*; Moscone, 1992). Particularly, in *S. elaeagnifolium* an euploid series has been cited (Moscone, 1992). All taxa analysed have comparatively small chromosomes as it is the rule in *Lycianthes* and *Solanum* (cf. Pijnacker & Ferwerda, 1984; Moscone, 1989a, 1992; Bernardello & Anderson, 1990; Bernardello & al., 1994). Nevertheless, we observe a maximum difference in the average chromosome length of 2.38-fold between species (see Table 2).

The karyotype analysis in *L. rantonnei* is the first reported in the genus. On the other hand, in the *Solanum* species here considered, only the karyotypes of *S. sisymbriifolium* (Badr & al., 1997) and *S. elaeagnifolium* (Patil, 1968) have been previously studied, although on different populations. Comparisons with our results are only tentative as other authors do not follow the widely accepted nomenclature of Levan & al. (1964). The lack of voucher specimen in the contribution by Patil (1968) introduces a further limitation. Main disagreements with our findings in the first species are the smaller chromosome size and the absence of NOR-bearing chromosomes in the descriptions by Badr & al. (1997), whereas in *S. elaeagnifolium*, Patil (1968) points out a majority of chromosomes with submedian centromere and two NOR-bearing pairs instead of one.

The presence of one chromosome pair with NORs

and attached satellites on short arms, as we find in the species herein studied, is the rule in diploid *Solanum* species (Magoon & al., 1962; Mitra, 1967; Wu & Li, 1985; Trivedi & Sinha, 1986; Okoli, 1988; Bernardello & Anderson, 1990; Bernardello & al., 1994). Remarkable exceptions are cited in *S. pseudolulo* Heiser and *S. basendopogon* Bitter, the former with two NOR-bearing pairs, one of them with NORs on long arms as in the latter species (Bernardello & Anderson, 1990; Bernardello & al., 1994). On the other hand, in the *S. indicum* L. complex a maximum of three NOR-carrying pairs has been reported (Krishnappa & Chennaveeraiah, 1975). In *Solanum* NORs are most frequently placed on sm chromosomes, although in sect. *Lasiocarpa* they are commonly in m chromosomes (cf. Bernardello & al., 1994). Furthermore, the comparatively large size of the NOR-bearing pair (among the six largest in the karyotype) is also a common phenomenon in the genus (Swaminathan, 1954; Patil, 1968; Krishnappa & Chennaveeraiah, 1975; Pijnacker & Ferwerda, 1984; Wu & Li, 1985; Trivedi & Sinha, 1986; Okoli, 1988; Bernardello & Anderson, 1990).

Although in the majority of the species here studied the appearance of satellites is regular, the range of occurrence among species varies between 50% and 100% of the cells, and sometimes they appear in only one homologue. This lack of constancy, also reported for other members of the family (Moscone, 1989a, b; Bernardello & al., 1994), could be the effect of pretreatment with spindle inhibitors (Suda, 1975), but satellites have been seen in metaphase plates with different degrees of chromosome contraction. The NOR expression in just one chromosome of the NOR-bearing pair is usually explained by the occurrence of nucleolar competition (differential amphiplasty) that takes place in interspecific hybrids (Pikaard, 1999), but none of the taxa examined are supposed to be of hybrid origin.

The present results point out a variation in satellite length both in *Lycianthes* and *Solanum*, with microsatellites being the most frequent type followed by macrosatellites. In this sense, a strikingly wide range of satellite size and, particularly, the presence of conspicuous linear satellites as observed in *S. euacanthum*, are unusual in the family (Moscone, 1989a). Polymorphism of satellites between individuals, cells, and even homologous NOR-bearing chromosomes of a single cell as found in most species here examined (67%) and also recorded in other Solanaceae (cf. Moscone, 1989a, Moscone & al., 1995), seems to be related to their heterochromatic constitution (Acosta, unpubl.). It is well known that ribosomal DNA and related highly repetitive sequences that make up the NOR-associated heterochromatin are prone to vary in number of copies. Otherwise, structural rearrangements including somatic chromosome mutations that affect only the satellites could be the mechanisms responsible for such polymorphism (cf. Sato, 1981).

Although we find variation in karyotype composition concerning both chromosome size and morphology, in general, the taxa studied display symmetrical karyotypes as it is characteristic in *Solanum* (cf. Moscone, 1989a; Bernardello & Anderson, 1990; Bernardello & al., 1994). Stebbins (1971) includes this genus in category 1A according to its high symmetry; however, our data show that most species here studied fall into type 2A such as several members of sect. *Basarthrum* (cf. Bernardello & Anderson, 1990). In respect to chromosome morphology, the revised karyotypes usually have a majority of m and sm chromosomes, with the former being more frequent, in agreement with previous findings in *Solanum* (Oinuma, 1949; Swaminathan, 1954; Magoon & al., 1962; Mitra, 1967; Krishnappa & Chennaveeraiah, 1975; Pijnacker & Ferwerda, 1984; Wu & Li, 1985; Okoli, 1988; Bernardello & Anderson, 1990; Bernardello & al., 1994; Badr & al., 1997). In this sense, the predominance of st chromosomes as we observe in *S. palinacanthum* is a novelty. Moreover, the absence of t chromosomes is typical of *Solanum*, where only *S. surattense* Burm. f. and two species of sect. *Basarthrum* are cited to show just one pair of this chromosome type (Trivedi & Sinha, 1986; Bernardello & Anderson, 1990). On the other hand, complements with rather homogeneous chromosome size, as we have described are the rule in *Solanum*. Bimodal karyotypes, as the one of *Solanum* (*Acanthophora*) sp. 2, have been heretofore not reported in the genus and are very rare in the whole family, i.e., only in a few species of *Capsicum* L. and *Nicotiana* L. including tobacco (cf. Goodspeed, 1954; Moscone, 1989a; Japan Tobacco Inc., 1994).

Our data on different infrageneric groups indicate that in *Solanum* as a whole, differences in karyotype asymmetry between species are seldom conspicuous (cf. Moscone, 1989a; Bernardello & Anderson, 1990; Bernardello & al., 1994). Thus, it seems that the great diversification in the genus has been associated with few chromosome rearrangements visible with conventional staining, i.e., large duplications, pericentric inversions, and reciprocal translocations of segments of unequal size. In this sense, cumulative small and cryptic structural changes have been proposed to play an important karyoevolutionary role in sections *Basarthrum* and *Lasiocarpa* (Bernardello & Anderson, 1990; Bernardello & al., 1994). It should be noted that linkage mapping studies based on molecular markers in *Solanum* sections *Petota* and *Etuberosum* have demonstrated the occurrence of several inversions and translocations during the evolution of these groups (Tanksley & al., 1992; Perez & al., 1999). A common karyotype pattern of homoge-

neous-sized chromosomes mostly with median and, sometimes, submedian centromeres, where difference in length is 1.40-fold between the largest and the smallest chromosome, appears widely distributed in *Solanum* (Mitra, 1967; Wu & Li, 1985; Okoli, 1988; Bernardello & Anderson, 1990; Bernardello & al., 1994; Badr & al., 1997). Therefore, a karyotypic orthoselection would have occurred in this genus as in other Solanaceae, which preserves rather similar complements, independently of the chromosome size, throughout a higher taxon because they seem to be more stable (cf. Brandham & Doherty, 1998; Moscone & al., 2003). Paracentric inversions, which do not affect chromosome morphology and are predicted to be the most likely form of chromosome rearrangements maintained evolutionarily, have occurred during the divergence of potato and tomato (*Lycopersicon esculentum* Mill.) as revealed by an RFLP mapping analysis (Bonierbale & al., 1988).

Karyotype data and systematics. — Karyotype features allow individual species to be distinguished. Thus, chromosome variation, although not always large, has accompanied evolutionary divergence of the taxa studied, a general phenomenon observed in both the plant and animal kingdoms (Goodspeed, 1954; Rieseberg, 2001). At the same time, karyotype similarities may indicate relationships between taxa as discussed in the following paragraphs.

Lycianthes in particular, is a taxon sometimes included in *Solanum* after Dunal (1852); however, several authors recognize its generic rank mainly on the basis of calyx vasculature and structure, which would indicate advanced status in relation to *Solanum* (cf. Bitter, 1919; D'Arcy, 1986; Nee, 1999; Hunziker, 2001). The distinction of *Lycianthes* at the generic level has been recently supported by molecular data on three species, which demonstrate its placement closest to *Capsicum* (Olmstead & Palmer, 1997; Bohs & Olmstead, 2001). In this context, our karyotype analysis of *L. rantonnei*, a shrub from subtropical forests (Barboza & Hunziker, 1992), does not show any major differences with the members of *Solanum* as a whole. At the same time, it stands separated from species of *Capsicum* by its smaller chromosome size (cf. Moscone, 1989a).

As very few species of *Solanum* subgen. *Solanum*, *Potatoe*, and *Brevantherum* have been considered in the present contribution, only preliminary conclusions on karyotype comparisons can be made. The three species of subgen. *Solanum*, growing in subtropical and temperate zones, exhibit different karyotype formulae but they have in common low A_2 asymmetry index values (see Table 2, Fig. 4). *Solanum chenopodioides*, a weedy annual herb included in a different section than the other members of this subgenus, which are perennial subshrubs (cf. Morton, 1976; Hunziker, 2001), is karyologically distinct in its smaller chromosomes and lower R value. On the other hand, *S. tucumanense* differs by having higher mean arm ratio and A_1 asymmetry index estimates. Recently, Knapp (2001) proposed that *S. tucumanense* Griseb. is a synonym of *S. pseudocapsicum* L.; nevertheless, according to Morton (1976) and Gutiérrez & Barboza (2003), both entities are clearly distinguished as independent species considering leaf and flower features and geographic distribution, with the former restricted to Paraguay and N Argentina and the latter spread as a weed in tropical regions throughout the world. Our data support the latter hypothesis as they show striking differences in karyotype formula, mean arm ratio and A_1 asymmetry index, with *S. tucumanense* having a majority of sm chromosomes and *S. pseudocapsicum* m ones. Further karyotype analyses in additional samples of both taxa are needed to assess the extent of these findings.

In subgen. *Potatoe*, all three species display similar A_1 asymmetry index estimates, but *S. palustre* (sub. nom. *S. brevidens* Phil.), a perennial herb growing at medium altitudes in temperate regions and placed into a separate section (cf. Morton, 1976; Contreras-M. & Spooner, 1999; Hunziker, 2001), has smaller chromosomes (see Table 2, Fig. 3). Otherwise, this species slightly differs in karyotype formula and NOR position from the perennial herbaceous *S. tripartitum* and the shrubby *S. endoadenium*, the latter having larger chromosomes and lower R value. It should be noted that the last two species have similar biogeographical patterns as they inhabit semiarid subtropical areas from low to high elevations. Lastly, *S. argentinum*, a subtropical shrub from low to medium altitudes belonging to subgen. *Brevantherum* (Morton, 1976; Hunziker, 2001), does not show any particular karyotypic feature that deviates from the typical karyotype pattern found in *Solanum* as a whole.

In the prickly *Solanum* subgen. *Leptostemonun*, the phenogram derived from karyotype features shows a species grouping that, in general, matches with the species arrangement in subsections proposed by Nee (1999) for the subgenus based on morphological characters (see Fig. 5). All species of sect. *Melongena* exhibit symmetrical complements, although both entities of subsect. *Cryptocarpum* (cluster 1 in the phenogram), which are weedy perennial shrubs with accrescent calyx and large prickles growing in humid subtropical areas of low to middle elevations, display a mean chromosome length twice as long as both entities of subsect. *Lathyrocarpum* (cluster 2 in part) (see Table 2, Fig. 4). The species examined of the latter subsection, although weedy, differ from those of the former subsection by being herbs with shorter prickles inhabiting arid subtropical and temperate zones, where they can reach higher altitudes (cf. Morton, 1976; Matesevach, 2002, pers. comm.). *Solanum eua-*

canthum, an annual with accrescent calyx, and *S. elaeagnifolium*, a perennial without calyx accrescence, are placed in different series of Nee's (1999) system, although they show similar karyotypes (phenetic dissimilarity value = 0.17) and can be distinguished from each other just by the NOR-bearing chromosome pair. Chromosome size differences have been of taxonomic value and used to delimit some litigious solanaceous groups, e.g., *Solanum* sects. *Cyphomandropsis* and *Pachyphylla*, both taxa with very large chromosomes in comparison to the remaining species of *Solanum*, which several authors consider, at least partly, as belonging to the independent genus *Cyphomandra* Sendtn. (cf. Roe, 1967; D'Arcy, 1972; Moscone, 1989a, 1992; Pringle & Murray, 1991; Bohs, 1994, 2001; Hunziker, 2001).

All members studied of sect. *Acanthophora* (clusters 2 in part, 3, and 4), 6 out of the 18 recognized species (Nee, 1999), are shrubs having long prickles, simple hairs, and winged seeds (except *S. palinacanthum*), which display considerable karyotype diversity with an increased asymmetry (except *S. atropurpureum*) (see Table 2, Figs. 3, 4). Species of subsect. 2, which grow in humid subtropical (sometimes also tropical) regions, have rather similar chromosome length and A_2 asymmetry index estimates. In the phenogram (see Fig. 5), these species are included together with species of subsect. *Lathyrocarpum* (cluster 2) in disagreement with Nee's (1999) sectional classification. In particular, *S. atropurpureum* and *S. tenuispinum* are species exomorphologically very close but quite different in their karyotypes (phenetic dissimilarity value = 0.72) and inhabiting disjunct areas, the former in Brazil, Paraguay, Uruguay, and NE Argentina at low to medium altitudes, and the latter, in Peru, Bolivia, and NW Argentina, where it reaches higher elevations (cf. Morton, 1976; Nee, 1991, 1999; Matesevach, 2002). Finally, *S. palinacanthum*, the only member examined of subsect. *Acanthophora*, and *Solanum* sp. 2, a species under taxonomic analysis and still without assigned subsection, both show the most asymmetrical and deviating karyotypes and, thus, they are isolated in the phenogram (clusters 3 and 4, respectively). The former is a weedy plant with large fruits, of extended subtropical geographic distribution from low to middle altitudes (Morton, 1976; Nee, 1991; Matesevach, 2002), while the latter is unique by having a calyx with large sepals of unequal size, small elliptic fruits, and a distribution restricted to humid regions of S Brazil and NE Argentina (Matesevach, pers. comm.).

In several plant groups, increased asymmetry is associated with advanced taxa (cf. Stebbins, 1971), and, in this case, it is postulated as an apomorphic condition. This could be applicable to our data on *Solanum* subgen. *Leptostemonum*, a group of probably ancient origin whose monophyly is proposed according to chloroplast DNA restriction site, and chloroplast *ndhF* and nuclear ITS DNA sequence analyses (Olmstead & Palmer, 1997; Bohs & Olmstead, 1999, 2001). Species of sect. *Acanthophora*, which generally show more asymmetrical karyotypes in comparison to species of sect. *Melongena*, also display some derived morphological features as flattened and winged seeds (in most cases, including *Solanum* sp. 2), long aciculate or narrow-based prickles, simple hairs in addition to stellate ones, and possibly, reduced calyx accrescence (cf. Morton, 1976; Whalen, 1984; Nee, 1991, 1999; Matesevach, 2002). In particular, *S. palinacanthum* carrying one of the most asymmetrical complements here observed, also possesses a strong tendency to andromonoecy and large berries, which are considered apomorphic states in the subgenus (Whalen, 1984). The anomalous chromosome number $x = 11$ reported in *S. mammosum*, a member of subsect. *Acanthophora*, provides further karyological evidence that sect. *Acanthophora* could be an advanced group in subgen. *Leptostemonum* (Madhavadian, 1968; Heiser, 1971). A broader karyotypic analysis together with a molecular phylogenetic study are needed in this subgenus and the genus as a whole, to gain a better knowledge of possible karyoevolutionary trends.

ACKNOWLEDGEMENTS

We are grateful to Prof. Ing. A. T. Hunziker, Dr. G. E. Barboza, and Biol. A. M. Matesevach for the identification of voucher specimens, and to several botanists who provided material. We are particularly in debt to G. E. Barboza and A. M. Matesevach for valuable suggestions and comments on the manuscript. M. C. Acosta acknowledges fellowship support of the National University of Córdoba (Argentina) for Ph. D. dissertation study and of the Latin-American Net of Botany for a study leave at the Department of Botany, Federal University of Pernambuco (Recife, Brazil). This work was supported by grants to E. A. Moscone from the National Council of Scientific and Technical Research (CONICET, Argentina) and the National University of Cordoba.

LITERATURE CITED

Badr, A., Khalifa, S. F., Aboel-Atta, A. I. & Abou-El-Enain, M. M. 1997. Chromosomal criteria and taxonomic relationships in the Solanaceae. *Cytologia* 62: 103–113.

Barboza, G. E. & Hunziker, A. T. 1992. Estudios sobre Solanaceae XXXIII. El género *Lycianthes* en la Argentina. *Darwiniana* 31: 17–34.

Battaglia, E. 1955. Chromosome morphology and terminology. *Caryologia* 8: 179–187.

Bernardello, L. M. & Anderson, G. J. 1990. Karyotypic studies in *Solanum* section *Basarthrum* (Solanaceae). *Amer. J. Bot.* 77: 420–431.

Bernardello, L. M., Heiser, C. B. & Piazzano, M. 1994. Karyotypic studies in *Solanum* section *Lasiocarpa* (Solanaceae). *Amer. J. Bot.* 81: 95–103.

Bitter, G. 1919. Die Gattung *Lycianthes*. *Abh. Naturwiss. Vereins Bremen* 24: 292–520.

Bohs, L. 1994. *Cyphomandra* (Solanaceae). *Fl. Neotrop. Monogr.* 63. The New York Botanical Garden, New York.

Bohs, L. 2001. Revision of *Solanum* section *Cyphomandropsis* (Solanaceae). *Syst. Bot. Monogr.* 61: 1–85.

Bohs, L. & Olmstead, R. G. 1999. *Solanum* phylogeny inferred from chloroplast DNA sequence data. Pp. 97–110 in: Nee, M., Symon, D. E., Lester, R. N. & Jessop, J. P. (eds.), *Solanaceae IV: Advances in Biology and Utilization.* Royal Botanic Gardens, Kew.

Bohs, L. & Olmstead, R. G. 2001. A reassessment of *Normania* and *Triguera* (Solanaceae). *Pl. Syst. Evol.* 228: 33–48.

Bonierbale, M. W., Plaisted, R. L. & Tanksley, S. D. 1988. RFLP maps based on a common set of clones reveal modes of chromosomal evolution in potato and tomato. *Genetics* 120: 1095–1103.

Bradley, M. V. 1948. A method for making aceto-carmine squashes permanent without removal of the cover slip. *Stain Technol.* 23: 41–44.

Brandham, P. E. & Doherty, M. J. 1998. Genome size variation in Aloaceae, an angiosperm family displaying karyotypic orthoselection. *Ann. Bot.* 82 (Suppl. A): 67–73.

Contreras-M., A. & Spooner, D. M. 1999. Revision of *Solanum* section *Etuberosum* (subgenus *Potatoe*). Pp. 227–245 in: Nee, M., Symon, D. E., Lester, R. N. & Jessop, J. P. (eds.), *Solanaceae IV: Advances in Biology and Utilization.* Royal Botanic Gardens, Kew.

D'Arcy, W. G. 1972. Solanaceae studies II: typification of subdivisions of *Solanum*. *Ann. Missouri Bot. Gard.* 59: 262–278.

D'Arcy, W. G. 1986. The calyx in *Lycianthes* and some other genera. *Ann. Missouri Bot. Gard.* 73: 117–127.

Dunal, M. F. 1852. Solanaceae. Pp. 1–690 in: De Candolle, A. P. (ed.), *Prodromus Systematis Naturalis Regni Vegetabilis*, vol. 13. V. Masson, Paris.

Edmonds, J. M. 1977. Taxonomic studies on *Solanum* section *Solanum* (Maurella). *Bot. J. Linn. Soc.* 75: 141–178.

Edmonds, J. M. & Chweya, J. A. 1997. *Black Nightshades:* Solanum nigrum *L. and Related Species.* IPGRI, Rome.

Ellis, R. H., Hong, T. D. & Roberts, E. H. 1985. *Handbook of Seed Technology for Genebanks.* 2. *Compendium of Specific Germination, Information and Test Recommendations. Handbook for Genebanks 3.* IBPGR, Rome.

Goodspeed, T. H. 1954. *The Genus* Nicotiana. Chronica Botanica Co., Waltham.

Guerra, M. 1983. O uso de Giemsa na citogenética vegetal: comparação entre a coloração simples e o bandeamento. *Ci. & Cult.* 35: 190–193.

Gutiérrez, M. A. & Barboza, G. E. 2003. Delimitation of *Solanum* sect. *Pseudocapsicum* in Argentina and the actual identity of *S. tucumanense* (Solanaceae). *Bol. Soc. Argent. Bot*. 38 (Suppl.): 91–92.

Hawkes, J. G. 1990. *The Potato: Evolution, Biodiversity and Genetic Resources.* Smithsonian Institution Press, Washington D.C.

Hawkes, J. G. 1999. The economic importance of the family Solanaceae. Pp. 1–8 in: Nee, M., Symon, D. E., Lester, R. N. & Jessop, J. P. (eds.), *Solanaceae IV: Advances in Biology and Utilization.* Royal Botanic Gardens, Kew.

Heiser, C. B. 1971. Notes on some species of *Solanum* (sect. *Leptostemonum*) in Latin America. *Baileya* 18: 59–65.

Howard, H. W. 1970. *Genetics of the Potato,* Solanum tuberosum. Springer-Verlag, New York.

Hunziker, A. T. 2001. *Genera Solanacearum: The Genera of Solanaceae Illustrated, Arranged According to a New System.* ARG Gantner Verlag K-G, Liechtenstein.

Infostat Group. 2002. *INFOSTAT . Version 1.1.* Facultad de Ciencias Agropecuarias, Universidad Nacional de Córdoba. Editorial Brujas, Argentina.

Japan Tobacco Inc. 1994. *The Genus* Nicotiana *Illustrated.* Japan Tobacco Inc., Tokyo.

Knapp, S. 2001. *Solanum* Section *Geminata* (Solanaceae). *Fl. Neotrop. Monogr.* 84. The New York Botanical Garden, New York.

Krishnappa, D. G. & Chennaveeraiah, M. S. 1975. Cytotaxonomy of *Solanum indicum* complex. *Cytologia* 40: 323–331.

Levan, A., Fredga, K. & Sandberg, A. 1964. Nomenclature for centromeric position in chromosomes. *Hereditas* 52: 201–220.

Madhavadian, P. 1968. Chromosome numbers in South Indian Solanaceae. *Caryologia* 21: 343–347.

Magoon, M. L., Ramanujam, S. & Cooper, D. C. 1962. Cytogenetical studies in relation to the origin and differentiation of species in the genus *Solanum* L. *Caryologia* 15: 151–252.

Matesevach, M. 2002. *Solanum* subgen. *Leptostemonum*. *Flora Fanerogámica Argentina*, fasc. 79: 1–35. Proflora CONICET, Cordoba.

Mitra, K. 1967. The significance of karyotype studies in relation to speciation in some solanaceous species. *Bull. Bot. Soc. Bengal* 21: 75–80.

Morton, C. V. 1976. *A Revision of the Argentine Species of Solanum.* Academia Nacional de Ciencias, Córdoba.

Moscone, E. A. 1989a. *Estudios Citotaxonómicos en las Tribus Solaneae y Nicotianeae (Solanaceae) de América del Sur.* Ph. D. dissertation, University of Córdoba, Argentina.

Moscone, E. A. 1989b. Karyotype analyses in three Patagonian and S. Andean endemic genera of Nicotianeae (Solanaceae). *Pl. Syst. Evol.* 166: 31–39.

Moscone, E. A. 1992. Estudios de cromosomas meióticos en Solanaceae de Argentina. *Darwiniana* 31: 261–297.

Moscone, E. A., Baranyi, M., Ebert, I., Greilhuber, J., Ehrendorfer, F. & Hunziker, A. T. 2003. Analysis of nuclear DNA content in *Capsicum* (Solanaceae) by flow cytometry and Feulgen densitometry. *Ann. Bot.* 92: 21–29.

Moscone, E. A., Loidl, J., Ehrendorfer, F. & Hunziker, A. T. 1995. Analysis of active nucleolus organizing regions in *Capsicum* (Solanaceae) by silver staining. *Amer. J. Bot.* 82: 276–287.

Nee, M. 1991. Synopsis of *Solanum* section *Acanthophora*: a group of interest for glycoalkaloids. Pp. 257–266 in: Hawkes, J. G., Lester, R. N., Nee, M. & Estrada, N. (eds.), *Solanaceae III: Taxonomy, Chemistry, Evolution.* Royal Botanic Gardens, Kew.

Nee, M. 1999. Synopsis of *Solanum* in the New World. Pp. 285–333 in: Nee, M., Symon, D. E., Lester, R. N. & Jessop, J. P. (eds.), *Solanaceae IV: Advances in Biology and Utilization.* Royal Botanic Gardens, Kew.

Ochoa, C. M. 1990. *The Potatoes of South America: Bolivia.* Cambridge Univ. Press, Cambridge.

Ochoa, C. M. 1999. *Las Papas de Sudamérica: Peru.* Parte I. Allen Press, Lawrence.

Oinuma, T. 1949. Cytological studies on the genus *Solanum* with special reference to the karyotypes. *Jap. J. Genet.* 24: 182–189.

Okoli, B. E. 1988. Cytotaxonomic study of five West African species of *Solanum* L. (Solanaceae). *Feddes Repert.* 99: 183–187.

Olmstead, R. G. & Palmer, J. D. 1997. Implications for the phylogeny, classification, and biogeography of *Solanum* from cpDNA restriction site variation. *Syst. Bot.* 22: 19–29.

Patil, R. P. 1968. The karyotype of *Solanum elaeagnifolium* Cav. *Curr. Sci.* 38: 142–143.

Perez, F., Menendez, A., Dehal, P. & Quiros, C. F. 1999. Genomic structural differentiation in *Solanum*: comparative mapping of the A- and E-genomes. *Theor. Appl. Genet.* 98: 1183–1193.

Pijnacker, L. P. & Ferwerda, M. A. 1984. Giemsa C-banding of potato chromosomes. *Can. J. Genet. Cytol.* 26: 415–419.

Pikaard, C. S. 1999. Nucleolar dominance and silencing of transcription. *Trends Pl. Sci.* 4: 478–483.

Pringle, G. J. & Murray, B. G. 1991. Karyotype diversity and nuclear DNA variation in *Cyphomandra*. Pp. 247–252 in: Hawkes, J. G., Lester, R. N., Nee, M. & Estrada, N. (eds.), *Solanaceae III: Taxonomy, Chemistry, Evolution.* Royal Botanic Gardens, Kew.

Randell, B. R. & Symon, D. E. 1976. Chromosome numbers in Australian *Solanum* species. *Austr. J. Bot.* 24: 369–379.

Rieseberg, L. H. 2001. Chromosomal rearrangements and speciation. *Trends Ecol. Evol.* 16: 351–358.

Roe, K. E. 1967. Chromosome size in *Solanum* and *Cyphomandra*: taxonomic and phylogenetic implications. *Amer. Naturalist* 101: 295–297.

Romero Zarco, C. 1986. A new method for estimating karyotype asymmetry. *Taxon* 35: 526–530.

Sato, S. 1981. Cytological studies on the satellited chromosomes of *Allium cepa*. *Caryologia* 34: 431–440.

Sneath, P. H. A. & Sokal, R. R. 1973. *Numerical Taxonomy: The Principles and Practice of Numerical Classification.* W. H. Freeman, San Francisco.

Snow, R. 1963. Alcoholic hydrochloric acid-carmine as a stain for chromosomes in squash preparations. *Stain Technol.* 38: 9–13.

Stebbins, G. L. 1971. *Chromosomal Evolution in Higher Plants*. E. Arnold Ltd., London.

Suda, Y. 1975. Somatic chromosome number and chromosomal morphology of *Anemone hepatica* L. var. *japonica* (Nakai) Ohwi. *Cytologia* 40: 693–711.

Swaminathan, M. S. 1954. Nature of polyploidy in some 48-chromosome species of the genus *Solanum*, section *Tuberarium*. *Genetics* 39: 59–76.

Tanksley, S. D., Ganal, M. W., Prince, J. P., de Vicente, M. C., Bonierbale, M. W., Broun, P., Fulton, T. M., Giovannoni, J. J., Grandillo, S., Martin, G. B., Messeguer, R., Miller, J. C., Miller, L., Paterson, A. H., Pineda, O., Röder, M. S., Wing, R. A., Wu, W. & Young, N. D. 1992. High density molecular linkage maps of the tomato and potato genomes. *Genetics* 132: 1141–1160.

Trivedi, R. N. & Sinha, A. K. 1986. Karyomorphological studies in three populations of *Solanum surattense*, a weed. *Cytologia* 51: 157–161.

Whalen, M. D. 1984. Conspectus of species groups in *Solanum* subgenus *Leptostemonum*. *Gentes Herb.* 12: 179–282.

Wu, S. & Li, Z. 1985. Preliminary studies of chromosome morphology of several wild and cultivated eggplants. *Acta Bot. Sin.* 27: 361–369.