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Caryological analysis of South American species of *Vernonia* (Vernonieae, Asteraceae)

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Abstract

In the present study 16 populations belonging to 13 species of the genus *Vernonia* Schreb. were examined cytologically. In total, six different chromosome numbers, which represent three basic numbers: $x = 10$, $x = 16$ and $x = 17$, were found. These results include the first chromosome number reports for the following four species: *V. lanifera* Cristóbal & Dematt. ($2n = 2x = 32$), *V. membranacea* Gardner ($2n = 2x = 34$), *V. salzmannii* DC. ($2n = 2x = 20$) and *V. scabrifoliata* Hieron. ($2n = 2x = 128$). Besides, a new chromosome number was found in *V. saltensis* Hieron. ($2n = 2x = 32$), for which only tetraploid populations have been previously recorded. The data obtained in this work, along with the information available from the literature, show that the genus *Vernonia* in South America is heterogeneous with basic chromosome numbers that range between $x = 9$ and $x = 19$. These numbers suggest that a combination of polyploidy and aneuploidy has played an important role in the evolution of the genus.

Keywords: *Chromosomes, cytology, evolution, taxonomy,* Vernonia

Introduction

The tribe Vernonieae is one of the largest groups of the Asteraceae with about 1700 species distributed in the tropical regions of Asia, Africa and America. It presents two major centers of diversification: one in southern Brazil, and the other in the tropical area of Africa (Jones 1979a).

Most of the Vernonieae species belong to the complex genus *Vernonia* Schreb., which in South America presents about 340–360 species mainly distributed in the north of Argentina, Brazil, Paraguay and Bolivia (Robinson 1999).

The Vernonieae are considered one of the most complex groups within the Asteraceae from a taxonomic viewpoint. Discussions have mainly centered around the delimitation of the huge genus *Vernonia* (Jones 1979a, 1981; Robinson 1999). The species of this group present a great variation in habit and morphology, which has contributed to the adoption of different criteria of taxonomic delimitation at the generic and infrageneric level (Baker 1873; Cabrera 1944; Keeley 1978; Jones 1979a, 1981). Recently, Robinson (1999) confined *Vernonia* to North America and redistributed all the South American species in 16 new genera. However, the new classification has not been adopted by many authors, because they have considered that the elevation of the traditional sections of *Vernonia* to generic level is still premature (Hind 1993; Keeley & Jansen 1994; Esteves et al. 2005).

Almost all the studies concerning the delimitation of infrageneric categories have always been based on external morphological features, such as inflorescence pattern, florets number, number and shape of the phyllaries, etc. (Gleason 1906, 1923; Cabrera 1944; Jones 1979a, 1981; Stutts 1988). Chromosome number has not been considered extensively in the proposed classifications. However, the cytological data show that the chromosomes are widely variable in number and morphology, which suggests that they can be useful in taxonomic and evolutionary studies (Keeley & Turner 1990; Ruas et al. 1991; Dematteis & Robinson 1997; Robinson 1999).

After a series of studies on Vernonieae chromosomes, Jones (1974, 1979b, 1982) suggested three

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basic numbers for *Vernonia*: $x = 9$ and $x = 10$ for Old World species, and $x = 17$ for the American taxa. In contrast, based on the available chromosome counts, Keeley and Turner (1990) proposed a large series of basic numbers which includes $x = 14$, $x = 15$, $x = 16$, $x = 17$ and $x = 19$ for the New World members of the genus. A more recent analysis indicated a large series of numbers ranging from $x = 9$ to $x = 19$ that would be the result of a complex combination of polyploidy and aneuploidy (Ruas et al. 1991).

The present paper reports original chromosome numbers of South American species of *Vernonia* and the cytological information is discussed in relation to the chromosome evolution of the genus.

Materials and methods

The specimens studied were obtained from natural populations growing at different locations of Argentina, Bolivia and Uruguay. The source of the examined specimens is presented in Table I. Voucher specimens are deposited at the herbarium of the Instituto de Botánica del Nordeste (CTES).

Mitotic chromosome preparations were made from root meristems obtained from germinating seeds. The rootlets were pretreated for about 3.5–4.5 h with 0.002 M 8-hydroxiquinoline solution at room temperature, fixed in 3:1 absolute alcohol–acetic acid during 24–72 h and then stained using Fuelgen's technique. In all samples at least twenty counts from 7 to 10 individuals were made to verify the observations.

Results

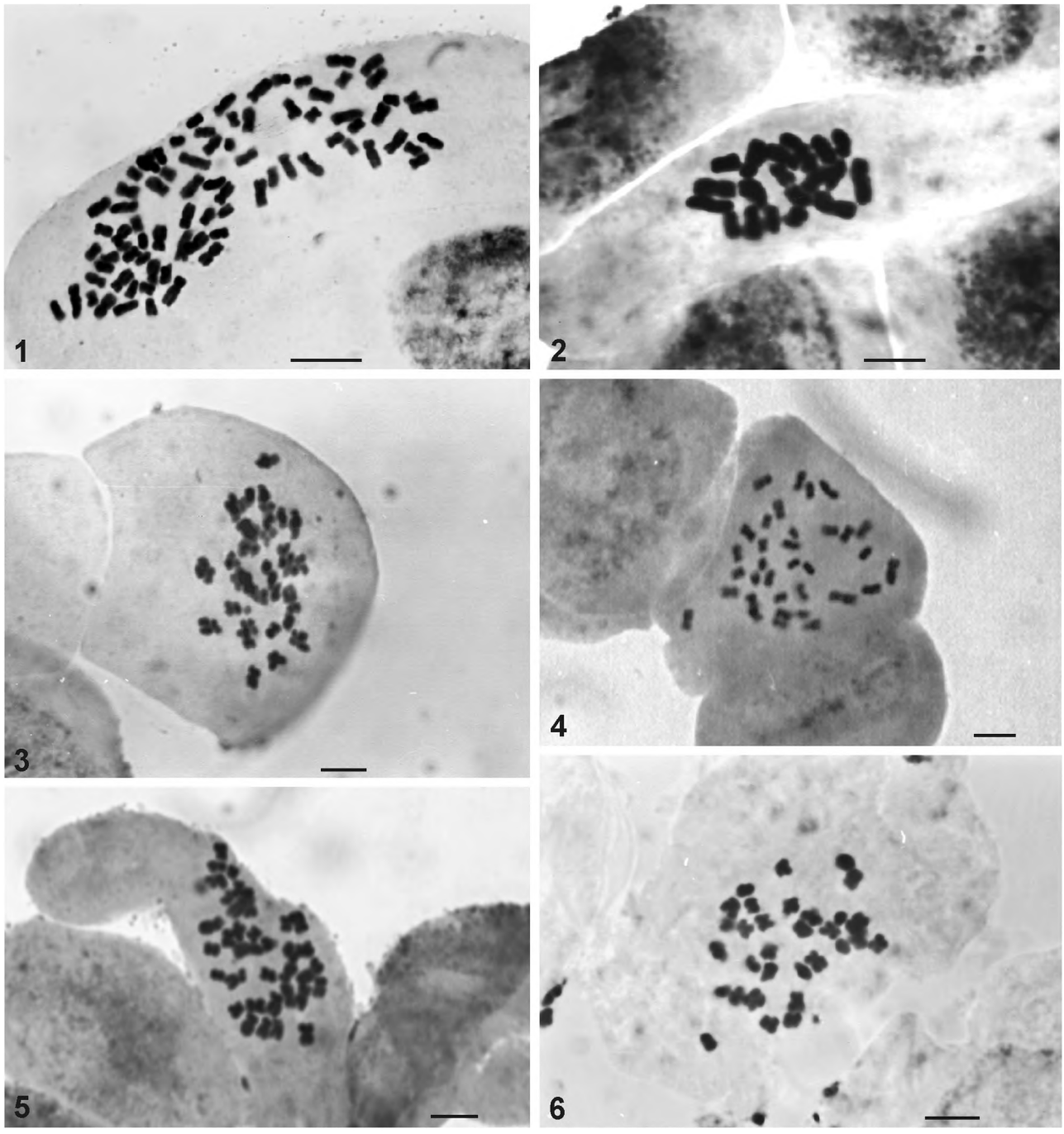
The analyzed species and their chromosome number are given in Table I. An asterisk (*) indicates that the species is studied for the first time, while a cross (+) shows that the chromosome number differs from those previously cited in the literature.

The chromosome numbers of 16 populations belonging to 13 species of *Vernonia* was determined. These include the first count for four species (*V. lanifera*, *V. membranacea*, *V. salzmanii* and *V. scabrifoliata*) and a new chromosome number for another taxon (*V. saltensis*). The chromosome counts

Table I. Examined specimens and chromosome numbers observed in 13 species of *Vernonia* from South America.

Species	2*n*	Voucher
V. amplexicaulis R.E.Fr.	$2n = 2x = 34$	Bolivia. Dept. Santa Cruz. Province Ñuflo de Chávez, 4 km E of San Javier, on the road to Concepción. *M. Dematteis et al. 1034* (CTES).
V. brasiliana (L.) Druce	$2n = 2x = 34$	Bolivia. Dept. Santa Cruz. Province Velasco, 1 km N of San Rafael. *M. Dematteis et al. 1059* (CTES).
V. cognata Less.	$2n = 8x = 80$	Argentina. Misiones. Dept. General Manuel Belgrano, Campina de Americo. *M. Dematteis & A. Krapovickas 1918* (CTES).
V. flexuosa Sims+	$2n = 2x = 20$	Uruguay. Dept. Lavalleja. 11 km S of Minas, on rocky soils. *M. Dematteis & A. Schinini 1671* (CTES).
V. lanifera Cristóbal & Dematteis*	$2n = 2x = 32$	Uruguay. Dept. Tacuarembó. Gruta de los Helechos, 15 km NW of Tacuarembó. *M. Dematteis & A. Schinini 1833* (CTES, SI).
V. lithospermifolia Hieron.	$2n = 2x = 20$	Argentina. Corrientes. Dept. Bella Vista. 10 km S of Bella Vista, Arroyo Toropí. *M. B. Angulo 8* (CTES).
V. loretensis Hieron.	$2n = 2x = 34$	Argentina. Corrientes. Dept. Mburucuyá, National Park Mburucuyá. *M. Dematteis et al. 1905* (CTES).
V. membranacea Gardner*	$2n = 2x = 34$	Bolivia. Dept. La Paz. Province Nor Yungas, 16 km N of Yolosa. *M. Dematteis et al. 1155* (CTES).
V. membranacea Gardner*	$2n = 2x = 34$	Bolivia. Dept. La Paz. Province Larecaja, 3 km N of La Aguada, between Guanay and Mapiri. *M. Dematteis et al. 1211* (CTES).
V. membranacea Gardner*	$2n = 2x = 34$	Bolivia. Dept. La Paz. Province Larecaja, 3 km N of La Aguada, between Guanay and Mapiri. *M. Dematteis et al. 1212* (CTES).
V. nudiflora Less.	$2n = 2x = 34$	Uruguay. Dept. Tacuarembó. Tacuarembó Chico river. *M. Dematteis & A. Schinini 1766* (CTES).
V. nudiflora Less.	$2n = 2x = 34$	Uruguay. Dept Rivera. 10 km S de Rivera, on the road to Tacuarembó. *M. Dematteis & A. Schinini 1479* (CTES, SI).
V. pinguis Griseb.	$2n = 4x = 68$	Bolivia. Dept. Tarija. Province Cercado, road from Papachacra toward Tipas. *F. Zenteno et al. 117* (CTES)
V. saltensis Hieron.+	$2n = 2x = 32$	Bolivia. Dept. Santa Cruz. Province Caballero, 5 km E of Saipina, on the road to Aiquile. *M. Dematteis et al. 2383* (CTES, SI).
V. salzmannii DC.*	$2n = 2x = 20$	Bolivia. Dept. La Paz. Province Larecaja, 3 km N of La Aguada, between Guanay and Mapiri. *M. Dematteis et al. 1210* (CTES, SI, LPB)
V. scabrifoliata Hieron.*	$2n = 8x = 128$	Bolivia. Dept. Santa Cruz. Province Velasco, 65 km E of Concepción, on road to San Ignacio. *M. Dematteis et al. 1046* (CTES).

+new chromosome number. *first chromosome count.



Figures 1–6. Mitotic chromosomes of *Vernonia*. Figure 1. *V. cognata*, $2n = 80$. 2. *V. lithospermifolia*, $2n = 20$. 3. *V. lanifera*, $2n = 32$. 4. *V. loretensis*, $2n = 34$. 5. *V. membranacea*, $2n = 34$. 6. *V. saltensis*, $2n = 32$. Scale bar = 5 μm.

realized in the remaining entities agree with previous records.

The chromosome numbers observed ranged between $2n = 20$ and $2n = 128$. Most of the examined species were diploids, but three polyploid entities were also found: one tetraploid with $2n = 4x = 68$ (*V. pinguis*) and two octoploids with $2n = 8x = 80$ and $2n = 8x = 128$ (*V. cognata* and *V. scabrifoliata*, respectively).

The analyzed species present three different basic numbers. *Vernonia amplexicaulis*, *V. brasiliana*, *V. loretensis* (Figure 4), *V. membranacea* (Figure 5), *V. nudiflora* and *V. pinguis* have the basic number $x = 17$. These species were mostly diploids with $2n = 34$, excepting *V. pinguis* that resulted tetraploid with $2n = 2x = 68$.

Other examined species, such as *V. cognata* (Figure 1), *V. flexuosa*, *V. lithospermifolia* (Figure 2)

and *V. salzmannii* have a basic number $x = 10$. Three of them were diploids with $2n = 20$, while *V. cognata* was octoploid with $2n = 80$.

The basic number $x = 16$ was found in three species: *V. saltensis* (Figure 6) and *V. lanifera* (Figure 3) were diploids with $2n = 32$, while *V. scabrifoliata* was octoploid having a chromosome number $2n = 128$.

Discussion

The basic number $x = 17$ was found in six species, five of them diploids and the remaining tetraploid. The same number has been previously determined in 24 other South American entities of the genus (Jones 1979b, 1982; Stutts 1988; Dematteis 2002) and the majority of species Central and North America (Keeley 1978; Jones 1979b). Most of the studies on *V. nudiflora* have reported $n = 17$ or $2n = 34$ (Jones 1974; Bernardello 1986; Stutts 1988; Ruas et al. 1991; Dematteis 1997, 2002). The single exception is the recount of $n = 16$ obtained by Covas and Hunziker (1954), which could be the result of intraspecific variation or an erroneous count.

The chromosome numbers found in *V. amplexicaulis* ($2n = 34$), *V. loretensis* ($2n = 34$) and *V. pinguis* ($2n = 68$) agree with the results of a previous analysis on specimens from Argentina (Dematteis 2002). For *V. brasiliana*, an earlier determination on Brazilian plants indicates $n =$ ca. 17 (Jones 1979b; sub *V. scabra* Pers.). No previous studies are available for *V. membranacea*, the remaining species having $x = 17$. Consequently, this constitutes the first chromosome count for this taxon.

Four examined species showed $x = 10$, a basic chromosome number that is relatively uncommon in the New World. The chromosome counts for *V. cognata* and *V. lithospermifolia* are in agreement with a previous study (Dematteis 1998), while in *V. salzmanii* ($2n = 2x = 20$) this constitutes the first cytological analysis for the species. In the remaining entity, *V. flexuosa*, the present one constitutes the second record of the diploid cytotype. Earlier studies indicate $n = 30$–32 in specimens of Buenos Aires (Hunziker et al. 1990) and $n = 20$ in plants of Brazil, Argentina and Uruguay (Ruas et al. 1991; Dematteis 1997, 2002). The latter populations are tetraploid, based on $x = 10$, while the material analyzed by Hunziker et al. (1990) could be an unstabilized hexaploid with this same basic number. The diploid cytotype has been previously found in plants of southern Paraguay (Dematteis et al. 2007), while our count has been made on specimens of central Uruguay.

Among the species with basic number $x = 16$, the chromosome counts realized here are the first for *V. lanifera* ($2n = 32$) and *V. scabrifoliata* ($2n = 128$), while in *V. saltensis* ($2n = 32$) a previous analysis reported $2n = 64$ (Dematteis 1998). The results obtained in this last species indicate that it has two different ploidy levels. The chromosome count of $2n = 64$ could represent a tetraploid population, while the sample analyzed here, having $2n = 32$, is diploid and constitutes the first report of that ploidy level for *V. saltensis*.

The present analysis, in addition to previous chromosome number reports, revealed that the genus is cytologically heterogeneous, particularly in South America, where basic numbers have been found ranging between $x = 9$ and $x = 17$, with $x = 16$ and $x = 17$ the most frequent (Keeley & Turner 1990; Dematteis 1997). In general, there is agreement that the basic chromosome number $x = 17$, present in many South American species, would have derived from $x = 9$ through duplication to 18 and subsequent reduction by aneuploidy (Jones 1979b; Turner 1981). According to this theory, it can be assumed that the other basic numbers observed in South American species, such as $x = 16$, $x = 15$ and $x = 14$, have been derived successively from $x = 17$ through aneuploidy or disploidy. These results corroborate the notion that polyploidy and aneuploidy have been of notable importance in the evolution of the New World Vernonieae.

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