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#### NOTHOSCORDUM MONTEVIDENSE SENSU LATO: NEW POLYPLOID CYTOTYPES IN ARGENTINA

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#### ABSTRACT

Two natural cytotypes of the *Nothoscordum montevidense* complex are described for the southeastern region of Buenos Aires province, Argentina. The cytotypes are 2n = 6x = 24 and 2n = 8x = 32 with metacentric chromosomes (m). Cytological and morphological evidence indicates that these cytotypes belong to *N. montevidense* ssp. *latitepalum*, although this subspecies is 2n = 4x = 16m. The probable origin of these cytotypes is discussed.

Key words: Alliaceae, Nothoscordum, polyploidy, polyploid cytotypes, autopolyploidy.

#### INTRODUCTION

In the Buenos Aires province the genus Nothoscordum Kunth, Alliaceae-Allieae (Dahlgren, Clifford, and Yeo 1985), contains seven species (Cabrera 1968). Guaglianone (1972) has divided the genus in two sections, Inodorum and Nothoscordum, the latter including the N. montevidense Beauv. complex: N. montevidense var. montevidense, N. montevidense var. minarum (Beauv.) Guagl., and N. montevidense var. latitepalum Guagl. Ravenna (1978) modified the taxonomic status of the varieties by placing them at the subspecies level.

The complex is distributed from Uruguay and Brazil to Argentina. In the latter country, *N. montevidense* s. l. occurs in lowland fields, in meadows, and on roadsides. The specimens are characterized by yellow flowers in several-flowered inflorescences (exceptionally one-flowered) with single bulbs or with bulblets and rhizomes. The plants grow in full sunlight and flower in spring and fall.

Two basic chromosome numbers have been reported, x = 4m and x = 5 (3m + 2t). Members of the genus are diploid, tetraploid, and hexaploid (Crosa 1972, 1974; Núñez, Frayssinet, Rodríguez, and Jones 1974) and are self-incompatible, except for the *N. inodorum* complex, which is apomictic (Núñez et al. 1974). Diploid and autotetraploid forms of *N. montevidense* s. l. occur in nature. The latter have, according to Núñez et al. (1974), a wider distribution.

The present paper deals with the description of polyploid populations belonging to the N. montevidense complex from the southeastern region of Buenos Aires province.

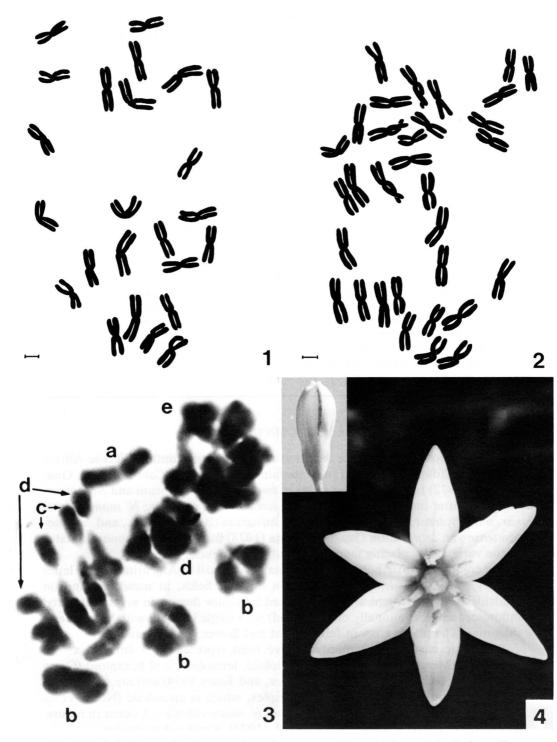


Fig. 1–4. 1–2. Mitoses showing metaphase chromosomes in two cytotypes of the *Nothoscordum* montevidense complex. -1, Cytotype I, 2n = 24m. -2. Cytotype II, 2n = 32m. -3. Meiotic pairing of cytotype I at first metaphase: a, monovalent; b, bivalents; c, trivalent; d, quadrivalents, and e,

Taxa	Source	Karyotypes (2n)	References Núñez et al. 1972		
var. montevidense	Buenos Aires (SI 26271)	16m			
var. montevidense	Uruguay	8m	Crosa 1972		
var. montevidense	Uruguay	16m	Crosa 1974		
var. <i>minarum</i>	Entre Ríos (Burk. 27914)	16m	Núñez et al. 1974		
var. <i>minarum</i>	Uruguay	10 (6m + 4t)	Crosa 1972		
var. <i>minarum</i>	Balcarce	10 (6m + 4t)	R. H. Rodríguez and A. E. Luchini (pers. comm.)		
var. <i>latitepalum</i>	Puán (SI 26544)	16m	O. Núñez and R. H. Rodrí- guez (unpublished data)		
ssp. latitepalum	Balcarce (Mon. 627)	24m	(1)		
(cytotype I)	Balcarce (Mon. 637)	24m	(1)		
	Cnel. Vidal (Mon. 639)	24m	(1)		
ssp. latitepalum	Balcarce (Mon. 637)	32m	(1)		
(cytotype II)	Balcarce (Mon. 645)	32m	(1)		
	Balcarce (Mon. 728)	32m	(1)		

Table 1. Karyotypes reported for the Nothoscordum montevidense complex.

(1): the authors in this paper.

#### MATERIALS AND METHODS

Four populations of the *N. montevidense* complex from Balcarce (627, 637, 645, 728) and one from Mar Chiquita (639) were analyzed. Plants of each locality were maintained in clay pots. Voucher specimens were deposited in the herbaria at Balcarce (BAL) and Buenos Aires (SI). For the proper determination of the plant material, morphological measurements of a total of 40 individuals from the five natural populations were made. The provenance and karyotypes of the examined material are listed in Table 1.

We determined chromosome numbers from root tips using the technique of Núñez et al. (1974). Meiotic analyses were made by squashing anthers, which previously had been fixed in alcohol: acetic acid (3:1), in acetic acid (45%) and staining them by following Snow's technique (Snow 1963). Drawings were prepared by the use of a Wild drawing apparatus and a Wild M-20 standard microscope. Chromosome types are indicated m and t in accordance with the nomenclature proposed by Levan, Fredga, and Sandberg (1964). Pollen fertility was estimated by using the technique proposed by Alexander (1969).

#### **RESULTS AND DISCUSSION**

#### Cytological Analysis and Fertility

We found natural populations of the *N. montevidense* complex with a somatic complement of 24 (cytotype I) (Fig. 1) and 32 m-chromosomes (cytotype II) (Fig. 2). On the basis of centromeric index (i), the m-chromosomes are metacentric and show a size gradation, with a mean index of 47 micrometers (i = short arm  $\times$  100/total chromosome length).

hexavalent. Arrows indicate a chain trivalent and a chain quadrivalent. -4. Upper view of a flower of cytotype I. The inset shows a closed flower with a streak on the outer tepal. (Fig. 1-2, scale = 10  $\mu$ m; Fig. 3, ×5000; Fig. 4, ×3.5 [inset, ×2]).

da la construcción de la	Number of <sup>2</sup>						
	1	2	3	4	5	6	
Total	36	612	12	68	2	25	
Average per cell	0.50	8.50	0.17	0.94	0.03	0.35	
Extreme value per cell	0–3	1-12	0-1	0–3	0-1	0-3	

Table 2. Frequency and distribution of associations at first metaphase of cytotype I.<sup>1</sup>

<sup>1</sup> Number of cells analyzed: 72.

<sup>2</sup> 1 = univalents, 2 = bivalents, etc.

In the genus *Nothoscordum*, meiosis occurs inside the bulb. This fact, as well as the one-flowered condition of these cytotypes, makes it difficult to analyze meiosis. For this reason, it has not been possible up to now to study meiosis in cytotype II. Chromosome pairing was analyzed at first metaphase in one clone of cytotype I (Fig. 3). The maximum pairing resulted in three hexavalents, one quadrivalent and one bivalent, and the minimum in twelve bivalents (Table 2). Because we observed a maximum pairing we consider that the chromosome complement is cytologically autohexaploid.

Both cytotypes are morphologically similar and so are their chromosomes. On this basis, it is postulated that cytotype II would be an autooctoploid.

Pollen from several plants of both cytotypes averaged 90% stainability. Seed set in selfed plants (hand-pollinated) averaged 0–3 seeds per fruit, whereas outcrossed plants averaged 15–20 seeds per fruit. Preliminary crosses between plants of cytotype I and cytotype II were successful, yielding up to 20 viable seeds per fruit.

#### Habitat and Morphology

Four of the studied populations grow in lowlands fields on flooded heavy clay soils, and one (728) on a hill of the Balcarce region (Table 1). In population 637, both cytotypes grow sympatrically.

Octoploid populations, which have been found in two widely disjunct regions, consisted of plants that were not morphologically separable from hexaploids in the same areas. Both cytotypes had one-flowered inflorescences (occasionally two-flowered under cultivation) with bright yellow flowers. The outer tepals were ovate with a purple streak on the outer face below (Fig. 4).

The cytotypes seemed to be closely related to ssp. *latitepalum* (Guaglianone 1972), the type specimen of which was tetraploid, 2n = 16m (Table 1).

Because of the short flowering period, the described cytotypes were hard to find in the field. Flowering occurs in full sun along with the flowering of *Oxalis chry*santha Prog. The cytotypes flowered from mid to late October along with ssp. *minarum*, also a yellow-flowered subspecies, while a white-flowered species, *N*. *bonariense* (Pers.) Beauv., 2n = 22m + 4t (Núñez, Frayssinet, and Rodríguez 1972), reached full flowering approximately a week later when both cytotypes and ssp. *minarum* had already dispersed their seeds.

Although the cytotypes and ssp. *minarum* present an overlapping flowering period, we did not find natural hybrids between them, but did observe plants (2n = 27m + 2t) intermediate to the cytotypes and *N. bonariense*. We also found one specimen with 2n = 28m from population 637 morphologically and cyto-

Cytotypes I and II	ssp. minarum			
x = 4m	x = 5 (3 m + 2t)			
1 scape	2-4 scapes			
Inflorescence 1-flowered, outer tepals 3.7 to 4.7 mm wide	Inflorescence 3-8-flowered, outer tepals up to 3 mm wide			
Bulb ovoid carrying bulblets and rhizomes	Bulb subglobose not carrying bulblets or rhi- zomes			

Table 3. Differential characteristics of the cytotypes and *Nothoscordum montevidense* ssp. *mina- rum*.

logically related to the cytotypes and probably descendent from a natural cross between 2n = 24 and 2n = 32.

Experimental crosses involving the various cytotypes within the *N. montevi*dense complex and *N. bonariense* are in progress.

Both cytotypes reproduce successfully by sexual and asexual means. The individuals have vegetative propagation and are capable of developing clonal patches which reach variable sizes within the dominant populations of diploid ssp. *minarum* and *N. bonariense*. Morphological differences between both cytotypes and sympatric ssp. *minarum* are shown in Table 3.

#### Hypotheses on the Origin of the Cytotypes

Studying the inheritance of B chromosomes, Luchini and Nicolini (pers. comm.) found two tetraploid and three triploid plants in the offspring of diploid ssp. *minarum*. The origin of the tetraploid cytotypes could be due to unreduced gametes in the diploid parents. Unreduced gametes with 10 chromosomes, fertilized by normally reduced gametes, would provide the complement of 15 chromosomes observed in the triploid hybrids.

The probable incidence of 2n gametes suggests a number of interesting possibilities for the origin of the cytotypes. Cytotype I could have originated from bilateral sexual polyploidization, BSP (Mendiburu and Peloquin 1976):  $2n = 16 \times 2n = 8$ , or from unilateral sexual polyploidization, USP:  $2n = 16 \times 2n = 16$ ; and cytotype II through BSP:  $2n = 16 \times 2n = 16$  or  $2n = 24 \times 2n = 8$ , or through USP:  $2n = 16 \times 2n = 24$ .

We have found no tetraploid plants with 2n = 16 in the study area. The discovery of this ploidy level would support the hypothesis about the origin of these cytotypes from 2n = 16m ancestors through sexual polyploidization. Also, we have found no diploids with 2n = 8m as reported by Crosa (1972). We conclude that cytotype I is autohexaploid and cytotype II is autooctoploid, based on the existence of 2n = 8m and 2n = 16m in the *N. montevidense* complex.

The described cytotypes confirm that polyploidization is an active mechanism in the evolution of the *N. montevidense* complex.

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