

## **A karyological study of *Allium rouyi* Gautier (Liliaceae), a recently rediscovered endemic species from the south of Spain**

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A population of *Allium rouyi*, previously considered an extinct species, has recently been rediscovered. From this sample, the somatic chromosome number and detailed chromosome morphology is presented. Using the karyological data, relationships between *A. rouyi* and allied species are discussed.

ADDITIONAL KEY WORDS:—Cytotaxonomy – Iberian Peninsula – karyogram.

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### INTRODUCTION

*Allium rouyi*, an endemic species from the south of Spain, was described by Gautier (in Rouy, 1898) from plants from Sierra Bermeja (Málaga). It has not been collected since until very recently, when a population of this taxon, considered extinct, was found among serpentine rocks of the Bermejense biogeographical sector, Mediterranean region (Sierra Bermeja, Estepona, Málaga), placed in the endemic vegetation series 'Pino pinastri–Quercetum cocciferae' (Cabezudo *et al.*, 1992). This species belongs to sect. *Scorodon* Koch, as shown by characters such as the membranous-fibrous tunic, usually filiform leaves, spathe with two short, unequal segments, campanulate perianth and simple

stamens. *A. rouyi* forms bulblets, unlike the other Iberian species of this section, *A. moschatum* L. and *A. chrysonemum* Stearn.

No previous karyological data for *A. rouyi* have been reported. In the present study, the chromosome number, karyograms, idiogram and asymmetry type are established for the first time.

#### MATERIAL AND METHODS

The material was cultivated in the greenhouse of the Department of Vegetal Biology and Ecology, University of Sevilla. The root tips of three bulbs were used for the study of chromosomes at mitosis. The roots were pretreated with 0.002M 8-hydroxyquinoline (Tjio & Levan, 1950) for 3–4 h at 4°C, fixed in Farmer's fluid (Löve & Löve, 1975) for 24 h and preserved in 70% ethanol. The root tips were stained in cold alcoholic-hydrochloric carmine solution (Snow, 1963). The material was squashed and mounted in 45% acetic acid. For chromosome morphology, the classification of Levan, Fregda & Sandberg (1964) has been followed. For apparent size of the chromosomes, the terminology of Pastor (1982) was used. Chromosomes between 2–5 µm are considered as small, between 5–9 µm as medium and over 9 µm as large. The karyotype asymmetry follows the terminology of Stebbins (1971). The idiogram was made from 15 somatic metaphases. Voucher specimens are lodged in the Herbarium of the Department of Vegetal Biology, University of Málaga (MGC) as follows: *Allium rouyi* Gautier. Spain: Málaga, Estepona, Sierra Bermeja, 9.vi.1992, Cabezudo, Pérez-Latorre, Navas & Recio (MGC 35000).

#### RESULTS

In the population studied, all the bulbs had a chromosome count of  $2n = 16$ , which corresponds to the diploid level with the basic number  $x = 8$ , and is the first count for this species. The apparent size of chromosomes varies between 6.4 and 15.2 µm (medium to large). The karyotype has a 1A type asymmetry and the karyogram (Fig. 1) presents seven pairs of metacentric chromosomes (m) and one pair submetacentric chromosomes (sm). The idiogram (Fig. 2) has the same formula as the karyogram: 14m+2sm.

#### DISCUSSION

On the basis of previous and present karyological data, the species from the Iberian Peninsula belonging to the sect. *Scorodon* (*A. moschatum*, *A. chrysonemum* and *A. rouyi*) have the basic number  $x = 8$ .

For *A. moschatum*, several authors have reported the somatic number  $2n = 16$ . Billeri (1954) and Garbari & Senatori (1975) found some pairs of submetacentric or subtelocentric chromosomes (sm-st), while Ozhatay (1983) reported the existence of metacentric chromosomes and one pair of submetacentric chromosomes. These authors showed the presence of satellited chromosomes in the karyotype of *A. moschatum*.

In *A. chrysonemum* the numbers  $2n = 32$  and 48 have been reported. The tetraploid level has been indicated by Stearn (1978), Ruíz Rejón, Lozano & Ruíz Rejón (1986) and Pastor & Valdés (1986), while the hexaploid level was



Figures 1 & 2. Chromosomes of *Allium rouyi* Gautier,  $2n = 16$ . Fig. 1 Karyogram. Fig. 2. Idiogram. Scale bar = 10  $\mu\text{m}$ .

found by Pastor (1985 sub. *A. reconditum*) and Pastor & Valdés (1986). The karyograms at both polyploid levels show subtelocentric chromosomes. In the tetraploid, chromosomes with satellites were sometimes observed (Pastor & Valdés, 1986). So far, no diploid counts have been reported for this species.

From morphological characters, as well as from the study of the pollen grains and seeds (Cabezudo *et al.*, 1992; Pastor, 1981), there is a close relationship between *A. rouyi* and *A. chrysonemum*. Nevertheless, the present karyological analysis reveals differences in the ploidy level, asymmetry and karyograms. So far, *A. rouyi* is only diploid ( $2x$ ), with  $2n = 16$ , while *A. chrysonemum* is both tetraploid ( $4x$ ) and hexaploid ( $6x$ ). The karyograms of *A. rouyi* have an asymmetry of type 1A and comprise 7 pairs of metacentric chromosomes (m) and one pair of submetacentric chromosomes (sm). No satellited chromosomes have been detected. Differing from *A. rouyi*, *A. chrysonemum* shows asymmetry type 2A, a karyogram with several pairs of subtelocentric chromosomes (st) and some chromosomes with satellites (Pastor 1985, Pastor & Valdés, 1986).

The results of this work reveal that to date *A. rouyi* is diploid only and has the most symmetrical karyotype of *Allium* section *Scorodon* in the Iberian Peninsula. Therefore, it may be considered to be less highly evolved than both *A. chrysonemum* and *A. moschatum*. Its condition of edaphic endemism (Nieto

Caldera, Pérez Latorre & Cabezudo, 1991), distributed in a very reduced and isolated area of the Bermejense sector (Sierra Bermeja, Málaga, Spain) could have contributed to this. For *A. chrysonemum*, its polyploid condition may help in the process of stabilization and establishment in new habitats, extending its area of distribution.

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