

Nuclear DNA content in Chilean species of *Phycella* and *Rhodolirium* (Amaryllidaceae)

Contenido de ADN nuclear en especies chilenas de *Phycella* y *Rhodolirium* (Amaryllidaceae)

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RESUMEN

Las determinaciones de ADN nuclear realizadas muestran que el valor 2C para *Rhodolirium montanum* Phil. ($2n=2x=16$) es de 17,39 pg y de 15,16 pg para *Phycella ignea* (Lindl.) Lindl. ($2n=2x=16$). Sin embargo, el tetraploide *Phycella scarlatina* Ravenna ($2n = 4x = 32$) tiene un valor de ADN 2C de 30,63 pg, el cual es concordante con su nivel de ploidía.

Angiosperms are the most intensively studied major group of organisms with published C-values varying about 2000-fold for a number over 4,400 studied species (Leitch *et al.* 2010). However, C-values for South American plants are scarce, which has been a preoccupation for researchers interested in study trends on genome size evolution of global floras (Bennett & Leitch 2005). In the case of Chilean angiosperms only 12 continental species have been studied and belong to seven families (ca. 0.24% of the Chilean angiosperms).

Rhodolirium and *Phycella* are two genera belonging to Amaryllidaceae that inhabit Chile. C-values have not been published for Chilean species of Amaryllidaceae, and only a previous contribution has been documented for species of the genera *Amaryllis* and *Hippeastrum* (Naranjo & Poggio 1988).

In this work the nuclear DNA content of *Rhodolirium montanum* Phil. [= *Rhodophiala rhodolirion* (Baker) Traub], *Phycella ignea* (Lindl.) Lindl., and *Phycella scarlatina* Ravenna from Central Chile are estimated for the first time, thus increasing the knowledge on 2C-values with data for two additional genera within Amaryllidaceae.

Plants of one accession of each species were collected from naturally growing populations. The voucher specimens were deposited at the herbarium from the Universidad de La Serena (ULS herbarium). The collection sites are shown in Table I. Measurements of nuclear DNA content (2C-values) were done by fluorescent microdensitometry in telophase nuclei of root tip cells obtained from five bulbs by each

species. The root tips were fixed in ethanol-glacial acetic acid (3:1 v/v) and stored in ethanol at 4°C. The root tips were stained using fluorescent Feulgen reaction (Prenna *et al.* 1974). The meristems were squashed onto glass slides, and the amount of fluorescence emitted by each telophase nucleus was measured using a Carl Zeiss Microscope Fluorometer (O'Neill *et al.* 1988). The fluorescent emissions (FE) contributed by each nucleus were estimated in arbitrary units. For the sampled species, FE values of 150 telophase (2C) nuclei were determined. The FE values in arbitrary units were converted into absolute mass of DNA (in picograms, pg) by comparison with root tip cells in telophase of *Allium cepa* ($2C = 33.5$ pg, $2n = 16$) (Johnston *et al.* 1999, Bennett & Leitch 2005).

Nuclear DNA content and chromosome numbers of *Rhodolirium montanum*, *Phycella ignea* and *P. scarlatina* are given in Table I. The higher 2C-value was estimated in *P. scarlatina* (30.6 pg) followed in decreasing order by *R. montanum* (17.3 pg) and *P. ignea* (15.1 pg). In all three cases the coefficient of variation was lower than 7.9%. The 2C-values of *R. montanum* and *P. ignea* are lower to the range reported for Amaryllidaceae which vary from 28 to ca. 164.3 pg (Leitch *et al.* 2005, 2010). A similar situation was observed comparing both species with the diploid taxa of *Hippeastrum* (range from 26.9 to 31.4 pg) (Naranjo & Poggio 1988). In the case of *P. scarlatina* its 2C-value is within the range described for Amaryllidaceae, being near to the recorded for some *Hippeastrum* species.

On the other hand, all the three examined species showed a basic chromosome number $x = 8$, where *R. montanum* and *P. ignea* have a chromosome number $2n = 2x = 16$, while *P. scarlatina* had $2n = 4x = 32$ (Palma-Rojas 2000, Muñoz *et al.* 2011). Interestingly, *P. scarlatina* doubled in $2n$ the number of both *P. ignea* and *R. montanum*, which is coincident with the doubling in $2C$ -value. These results corroborate the preliminary data described by Palma-Rojas

(2000) for the same species who suggested tetraploidy for *P. scarlatina* (likely allotetraploidy).

In Chile, ca. 43 species are recognized within Amaryllidaceae and belong to seven genera (Hoffmann 1989). Thus, $2C$ -values have been estimated only for ca. 6.9% of the Chilean species. Then, additional studies need to be carried out to complete the knowledge on genome size within this native family.

TABLE I. Collection sites, chromosome number and $2C$ DNA content for the examined *Rhodolirium* and *Phycella* species. FE, fluorescence emission in arbitrary units; SD, standard deviation.

TABLA I. Sitios de colecta, número cromosómico y contenido $2C$ de ADN para las especies examinadas de *Rhodolirium* y *Phycella*. FE, emisión de fluorescencia en unidades arbitrarias; SD, desviación estándar.

TAXA	COLLECTION SITES	$2n$	FE	$2C$ -VALUE (pg) MEAN \pm SD
<i>Rhodolirium montanum</i>	Metropolitana. Prov. Santiago, La Parva, altitude 2,683masl, (33°20'S- 70°17'W). 29-I-2009. C.Palma.	16	34.78	17.3 \pm 1.27
<i>Phycella ignea</i>	Metropolitana. Prov. Santiago, Loma Las Burras, altitude 531masl (33°27'S-70°50'W). 24-X-2009. C.Palma.	16	30.32	15.1 \pm 1.20
<i>P. scarlatina</i>	Coquimbo. Prov. Elqui, Quebrada El Arrayán, altitude 108masl (29°49'S; 71°07'W). 05-IX-2009. C.Palma.	32	61.26	30.6 \pm 1.79

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BIBLIOGRAPHY

- BENNETT, M. & I. LEITCH. 2005. Nuclear DNA amounts in Angiosperms: Progress, problems and prospects. *Annals of Botany* 95: 45-90.
- HOFFMANN, A. 1989. Chilean monocotyledonous geophytes taxonomic considerations and their state of conservation. *Herbertia* 45(1): 14-28.
- JOHNSTON J.S., M.D. BENNETT, A.L. RAYBURN, D.W. GALBRAITH & H.J. PRICE. 1999. Reference standards for determination of DNA content of plant nuclei. *American Journal of Botany* 86: 609-613.
- LEITCH, I., D. SOLTIS, P. SOLTIS & M. BENNETT. 2005. Evolution of DNA amounts across land plants (Embryophyta). *Annals of Botany* 95: 207-217.
- LEITCH, I., J. BEAULIEU, M. CHASE, A. LEITCH & M. FAY. 2010. Genome size dynamics and evolution in Monocots. *Journal of Botany* 2010: 18 pp. URL: <http://www.hindawi.com/journals/jb/http://www.hindawi.com/journals/jb/2010/862516/cta/> (accessed May 28, 2011).
- MUÑOZ, M., R. RIEGEL, P. SEEMAN, P. PEÑAILILLO, F. SCHIAPPACASSE & J. NÚÑEZ. 2011. Relaciones filogenéticas de *Rhodolirium montanum* Phil. y especies afines, basadas en secuencias nucleotídicas de la región ITS y análisis cariotípico. *Gayana Botánica* 68(1): 40-48.
- NARANJO, C. & L. POGGIO. 1988. A comparison of karyotype, Ag-NOR bands and DNA content in *Amaryllis* and *Hippeastrum* (Amaryllidaceae). *Kew Bulletin* 43: 317-325.
- O'NEILL, P., M.B. SINGH & R.B. KNOX. 1988. Cell biology of the stigma of *Brassica campestris* in relation to CO₂ effects on self-pollination. *Journal of Cell Science* 89: 541-549.
- PALMA-ROJAS, C. 2000. Caracterización citogenética de los géneros *Rhodophiala* Presl. y *Phycella* Lindl. (Amaryllidaceae). En: P. Peñailillo, F. Schiappacasse (eds.), Los geófitos nativos y su importancia en la floricultura: Fundación para la Innovación Agraria (FIA) y Dirección de Investigación, Universidad de Talca (DIUT), Santiago, Chile, pp. 73-79.
- PRENNA, R., S. LEIVA & G. MAZZINI. 1974. Quantitation of DNA by cytofluorometry of the conventional Feulgen reaction. *Histochemical Journal* 6: 467-484.

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