# A KARYOLOGICAL INVESTIGATION ON SOME RARE AND ENDANGERED SPECIES OF AMARYLLİDACEAE İN TURKEY

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#### Abstract

Chromosome number and morphology of three species belonging to Amaryllidaceae viz., Galanthus rizehensis Stern., Leucojum aestivum L., and Pancratium maritimum L., from the Central Black Sea Region of Turkey were analyzed. The chromosome number of P. maritimum and L. aestivum is 2n=22 while of G. rizehensis it is 2n=26.

#### Introduction

The number of some plant species is gradually decreasing due to human and industrial effects. According to Ekim *et al.*, (2000) *Galanthus rizehensis* and *Leucojum aestivum* (Amaryllidaceae) are classified as vulnerable (V) while *Pancratium maritimum* is endangered (En). Distribution centers of *Galanthus* are in Anatolia, the Caucasus and in the Black Sea Region. *Galanthus* includes a lot of species in the Eastern Mediterranean. *G. rizehensis* is a perennial, bulbous plant often used as ornamental and in the treatment of some disease (Kutbay & Kılınç, 1995). *L. aestivum* is the only member of its genus distributed in Thrace and Northern Anatolia in Turkey. This species is a bulbous plant with very important medicinal uses, because of its rich alkoloid content (Kutbay & Kılınç, 1993). *P. maritimum* is the only member of *Pancratium* widely distributed along the Black Sea and Mediterranean Coasts of Turkey. *P. maritimum* also a bulbous plant is used for decoration and for medicinal purposes (Kılınç & Yüksel, 1995).

The present report describes the number and morphological properties of chromosomes in Turkish populations of these G. *rzehensis, L. aestivum* and *P. maritimum* which are decreasing rapidly in their frequency in the recent years.

### Material and Methods

Herbarium specimens of *G. rizeheńsis*, *L. aestivum* and *P. maritimum* were collected in the vicinity of Alaçam, Çakırlar copse and Kızılay camp (A6 grid square). Specimens are deposited in the herbarium of Ondokuz Mayıs University, Faculty of Arts and Science, Department of Biology and numbered, 150,153 and 157, recpectively.

The cytological investigation was carried out by means of root-tip squashes according to Özyurt *et al.*, (1996). Root tips of all the three species were collected from in their natural habitat in Northern Anatolia of Turkey. They were pretreated with saturated solution of  $\propto$  monobromonaphtalene (16h) and fixed in a mixture of ethanol and acetic acid (1h). Root tips were hydrolyzed with 1 N HCl for 10 minute at 60°C in an oven, then stained with feulgen reagent for 1 hour in darkness and finally squashed in 45% acetic acid. Slides were examined under a Nikon photomicroscope and photographs were taken with the same microscope.

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The karyograms were drawn from mitotic metaphase. Measurements were made on each pairs of mitotic chromosomes. Karyotype analysis were carried out according to the method described by Naranjo *et al.*, (1986) and by Levan *et al.*, (1964).

## Results

**1.** *Galanthus rizehensis*: The chromosome number of this species was 2n=26 (Fig. 1). The karyotype of this species consisted of 3 pairs of submedian (sm), 6 pairs of median (m) and 4 pairs of subterminal (st) (Fig. 2). The 1<sup>st</sup>, 2<sup>nd</sup>, 9<sup>th</sup>, 11<sup>th</sup>, 12<sup>th</sup>, 13<sup>th</sup> chromosomes are median, the 4<sup>th</sup>, 8<sup>th</sup>, 10<sup>th</sup>, chromosomes are submedian, 3<sup>rd</sup>, 5<sup>th</sup>, 6<sup>th</sup> and 7<sup>th</sup> chromosomes are subterminal centromeres (Table 1). Satellite is present on the short arm of the 7<sup>th</sup> chromosome. Chromosome length range from 5.5 to  $20\mu$ . Longest arm is  $10\mu$  and shortest arm is  $2\mu$ . Karyotype details including chromosome length and arm ratio are shown table 1.

**2.** *Leucojum aestivum*: The chromosome number of this species was 2n=22 (Fig. 3). The karyotype of this species consisted of 1 pair of median, 1 pair of submedian and 8 pairs of subterminal chromosomes. The  $1^{st}$  is median, the  $8^{th}$  is submedian and other are subterminal centromeres (Fig. 4).

No satellite were present in the karyotype of this species. Chromosome lengths range from 6.5 to  $20\mu$ . The longest arm is 12  $\mu$  and the shortest is 1  $\mu$  (Table 2).

**3.** *Pancratium maritimum*: The chromosome number of this species was 2n=22 (Fig. 5). The karyotype of this species consist of 5 pairs of median (m) and 5 pairs of subterminal (st) and 1 pair of submedian (sm) chromosomes. The  $1^{st}$ ,  $2^{nd}$ ,  $3^{rd}$ ,  $4^{th}$  and  $6^{th}$  are median,  $10^{th}$  is submedian while the others have subterminal centromeres (Fig. 6). No satellites were present in the karyotype of this species. Chromosome total lengths range from 6 to  $11.5 \mu$ . The longest arm is  $6\mu$  and shortest is  $1\mu$ . The mean length and arm ratio of each of the chromosomes are presented in Table 3.

## Discussion

We determined the chromosome number of G. rizehensis as 2n=26 and P. maritimum and L. aestivum as 2n=22. The present study supports the contention presented by D'amato & Bianchi (1999) that the chromosome numbers for the L. aestivum are 2n=22 in Italian population. The karyotype comprises of one long pair of metacentric (m) and 10 medium sized pairs of st chromosomes two st pairs were satellited. In our observation this species has one pair m, 9 pairs st and one pair st-sm chromosomes. Satellite was not observed this species. Our findings are similar to the reports of Love (1973) who found that the chromosome number of L. aestivum which is distributed in Yugoslavia is 2n=22. D'amato & Bianchi (1999) reported that G. nivalis showed 2n=24 chromosomes, the set consisted of two long metacentric pairs with seven sm-st medium size chromosome pairs and three median pairs. In our study we observed that G. rizehensis has 2n=26 chromosomes, the set consists of two submedian pairs, seven median pairs, four subterminal pairs. This species has satellite on the 7th chromosomes. Chromosome number of G. rizehensis is given as 2n=24-36 in Flora of Turkey (Davis, 1984). The cytological studies in Galanthus have revealed a polimorphism among different karyotypes too and SAT chromosomes have been reported to be important in the

Table 1. Karybrype details of somatic enfomosomes of Outaninus recentisis								
Chromosome	С	L	S	L/S	SAT	I	Centromeric	
pairs	( <b>µ</b> )	( <b>µ</b> )	(μ)				position	
1	20	10	10	1,00	-	50	Median region (M)	
2	16,5	9,5	7	1,35	-	42	Median region (m)	
3	14,5	12	2,5	4,80	-	17	Subterminal(st)	
4	12	9	3	3,00	-	25	Submedian(sm)	
5	11	8,5	2,5	3,40	-	22	Subterminal(st)	
6	10,5	8	2,5	3,20	-	23	Subterminal (st)	
7	10	8	2	4,00	*	20	Subterminal (st)	
8	10	7,5	2,5	3,00	-	25	Submedian(sm)	
9	8	4,5	3,5	1,28	-	43	Median region(m)	
10	6	4	2	2,00	-	33	Submedian region	
11	6	3	3	1,00	-	50	Median region	
12	5,5	3	2,5	1,20	-	45	Median region	
13	5,5	3	2,5	1,20	-	45	Median region	

Table 1	. Karvotype	details of	somatic	chromosomes	of	Galanthus	rizehensis.
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Table 2. Karyotype details of somatic chromosomes of *Leucojum aestivum*.

Chromosome	С	L	S	L/S	SĀT	Ι	Centromeric
pairs	( <b>µ</b> )	( <b>µ</b> )	(μ)				position
1	20	10	10	1,00	-	50	Median region (M)
2	14	12	2	6,00	-	14	Subterminal (st)
3	13,5	10,5	3	3,50	-	22	Subterminal (st)
4	12,5	11,5	1	11,50	-	8	Subterminal (st)
5	10	. 8	2	4,00	-	20	Subterminal (st)
6	10	8	2	4,00	-	20	Subterminal (st)
7	9,5	8	1,5	5,33	-	15	Subterminal (st)
8	8	6	2	3,00	-	25	Submedian(sm)
9	8	6,5	1,5	4,33	-	18	Subterminal (st)
10	7,5	6,5	1,5	2,10	-	20	Subterminal (st)
11	6,5	5,5	1	5,00	-	15	Subterminal (st)

	Table 3. Kar	ryotype details of	somatic cl	hromosomes of	Pancratium n	ıaritimum.
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Chromosome	C	L	S	L/S	SAT	Ī	Centromeric
pairs	( <b>µ</b> )	( <u>µ</u> )	( <b>µ</b> )				position
1	11,5	6	5,5	1,09		47	Median region(m)
2	11,5	6	5,5	1,09	-	47	Median region(m)
3	10	5	5	1	-	50	Median region(M)
4	9,5	5	4,5	1,11	-	47	Median region(m)
5	7	5,5	1,5	3,66	-	21	Subterminal(st)
6	7	4	3	1,33	-	42	Median region(m)
7	6,8	5,8	1	5,8	-	14	Subterminal(st)
8	6,8	5,8	1	5,8	-	14	Subterminal(st)
9	6,7	5,7	1	5,7	-	-14	Subterminal(st)
10	6,2	4,2	2	2,1	-	32	Submedian (sm)
11	6	5	1	5	-	16	Subterminal(st)

C: Total chromosome length, S: Short arm length, SAT: Satellite, I: Centromeric index L: long arm length, L/S: Arm ratio



Fig. 1. Microphotograph of somatic metaphase chromosomes of G. rizehensis.



Fig. 2. Idiogram of chromosome complement of G. rizehensis of mitotic metaphase.



Fig. 3. Microphotograph of somatic metaphase chromosomes of L. aestivum



Fig. 4. Idiogram of chromosome complement of L. aestivum at mitotic metaphase.



Fig. 5. Microphotograph of somatic metaphase chromosomes of *P. maritimum*.



Fig. 6. Idiogram of chromosome complement of P. maritimum at mitotic metaphase.

variation (Zeybek, 1988). As a result of the observation given above we found the satellite is present on 7<sup>th</sup> of *G. rizehensis* karyotypes. D'amato & Bianchi (1999) reported that the occurence of accessory chromosomes is very common in *Galanthus* but we have not observed this chromosomes in *G. rizehensis*. The karyotypes of three species are more or less similar with respect to centromere position. The karyotypes are less specialized and predominated by metacentric (m) and subtelocentric (st) chromosomes while submetacenric (sm) are rare and acrocentric and telocentrics are totally absent. Moreover the karyotype of *G. rizehensis* is distinctive from others in as much as pair of SAT chromosomes is present in this. A comparative analysis of the chromosome morphology of the three species reported here shows that the chromosome are generally large in size (ranging in length between 5.5 and  $20\mu$ ). It is generally assumed that *Galanthus* and *Leucojum* represent to strictly related genera sharing many morpho-ecological traits(D'amato & Bianchi, 1999). The karyotype features of these genera could be correlated with the external morphological characteristics.

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