

KARYOTYPE ANALYSIS OF *MINUARTIA MESOGITANA* SUBSP. *MESOGITANA* AND *MINUARTIA ELMALIA* (CARYOPHYLLACEAE, ALSINOIDEAE)

OĞUZ YÜCE¹, HALİL ERHAN EROĞLU^{2*}, MURAT KOÇ² and ERGIN HAMZAOĞLU³

Summary: Mitotic metaphase chromosomes, karyotypic characters, monoploid karyograms and ideograms of *Minuartia mesogitana* subsp. *mesogitana* and *Minuartia elmalia* were investigated. Analysis of somatic metaphases showed that the chromosome numbers and the karyotype formula of these taxa were $2n = 2x = 24 = 8m + 8sm + 8st$ for *Minuartia mesogitana* subsp. *mesogitana* and $2n = 2x = 32 = 12m + 16sm + 4st$ for *Minuartia elmalia*. No satellites were observed in the karyotypes of the taxa. The intrachromosomal and interchromosomal karyotype asymmetries were estimated with Mean Centromeric Asymmetry (M_{CA}) and Coefficient of Variation of Chromosome Length (CV_{CL}).

Key words: *Minuartia*, karyotype, ideogram, karyotype asymmetry.

Resumen: Análisis cariotípico de *Minuartia mesogitana* subsp. *mesogitana* y *Minuartia elmalia* (Caryophyllaceae, Alsinoideae). En este trabajo se analizan los cromosomas metafásicos mitóticos, cariotipo, cariogramas e idiogramas de *Minuartia mesogitana* subsp. *Mesogitana* y *Minuartia elmalia*. El análisis de metafases somáticas mostró que el número cromosómico y la fórmula cariotípica de estos taxones fueron $2n = 2x = 24 = 8m + 8sm + 8st$ para *Minuartia* subsp. *Mesogitana* y $2n = 2x = 32 = 12m + 16sm + 4st$ para *Minuartia elmalia*. No se observaron satélites en los cariotipos de los taxones. La asimetría intracromosómica e intercromosómica fue estimada con la asimetría centroméricas promedio (MCA) y el coeficiente de variación de longitud cromosómica (CVCL).

Palabras clave: *Minuartia*, cariotipo, idiogramas, asimetría.

INTRODUCTION

The genus *Minuartia* L. is placed in the subfamily Alsinoideae (DC.) Fenzl of the Caryophyllaceae. It consists of annual or perennial herbaceous approximately 120 species found in the Northern Hemisphere, Argentina, Brazil and Chile (Bittrich, 1993; Nicola & Pozner, 2013). The genus especially perennial ones grow in extreme habitat conditions such as alpine, stony soils and rocky places (McNeill, 1962, 1963). It was reported that the *Minuartia* is a polyphyletic genus and should be

separated to at least three subgenera (Harbaugh *et al.*, 2010; Greenberg & Donoghue, 2011).

Different chromosome counts have been reported in the *Minuartia* till now. However, the most frequent chromosome number in the *Minuartia* is $2n = 2x = 30$. Other counts such as $2n = 2x = 14, 18, 20, 22, 24, 26, 28, 60$ and 90 are also known (Çelebioglu & Favarger, 1983, 1984, 1990; Davis *et al.*, 1988; Kamari *et al.*, 1996; Tan & Vural, 2000; Eroğlu *et al.*, 2013). The diploid chromosome number of *Minuartia mesogitana* Hand.-Mazz. subsp. *mesogitana* is not clear certain and was reported as $2n = 22, 24$ (Çelebioglu & Favarger, 1983, 1984, 1990; Kamari *et al.*, 1996). Also, chromosomal measurements of the *Minuartia mesogitana* subsp. *mesogitana* are not known. The aim of this study is to investigate the chromosome number, karyotype, ideogram and other detailed measurements of *Minuartia mesogitana* subsp. *mesogitana* and *Minuartia elmalia* (Aytaç) Aytaç, Parolly & Eren.

¹ Bozok University, Science Institute, Department of Biology, Yozgat, Turkey

² Bozok University, Faculty of Science and Art, Department of Biology, Yozgat, Turkey

³ Gazi University, Faculty of Education, Department of Primary School Teaching, Ankara, Turkey

* herhan.eroglu@bozok.edu.tr; heeroglu@yahoo.com

Table 1. Collection data of the studied taxa.

Taxa	Locality	Coordinates
<i>Minuartia mesogitana</i> subsp. <i>mesogitana</i>	TURKEY. Kayseri: Pınarbaşı, between Aşağıbeyçayır village and Yukarıbeyçayır village, 1755 m, 25-VI-2010, Hamzaoğlu 5699 & Koç (BOZOK)	38° 39' 23" N; 36° 27' 12" E
<i>Minuartia elmalia</i>	TURKEY. Antalya: Elmalı, Bey mountain, Küçüksöğle village, 2240 m, 28-VII-2012, Hamzaoğlu 6548 & Koç (BOZOK)	36° 37' 07" N; 30° 08' 00" E

MATERIAL AND METHODS

Collection information of the plant taxa are given in Table 1. The seeds were germinated between moist Whatman papers in Petri dishes. The root tips were fixed in Carnoy's fixative (3:1 absolute alcohol: glacial acetic acid) at +4°C overnight after pre-treatment in colchicine (0.2% w/v) for 2 hours. Root tips were stained with acetocarmine. Preparations were made using the squash method. At least ten metaphase cells were used to determine chromosome numbers. The metaphase chromosomes were photographed by Olympus BX53 digital camera and measured with the Bs200ProP image processing and analysis system.

Karyotype formula was determined by chromosome morphology based on centromere position according to Levan classification (Levan *et al.*, 1964). The following parameters were estimated to characterize the karyotypes numerically: long arm (l), short arm (s), total chromosome length (TCL) = [l + s], arm ratio (r) = [l / s] and centromeric index (CI) = [s / (l + s) × 100]. For each taxon, karyograms were drawn based on length of chromosome size (arranged large to small).

The interchromosomal and intrachromosomal karyotype asymmetries were estimated with mean centromeric asymmetry (M_{CA}) and coefficient of variation of chromosome length (CV_{CL}), both indices varies between 0 (perfectly symmetric, no variation) and 100 (perfectly asymmetric, total variation) (Paszko, 2006; Peruzzi & Eroğlu, 2013). Asymmetry indexes were compared with Pearson correlation.

RESULTS

Minuartia mesogitana subsp. *mesogitana*

Mitotic metaphase chromosomes, karyogram and monoploid ideogram of *Minuartia mesogitana* subsp. *mesogitana* are given in Fig. 1A. The measurement data of these chromosomes are given in Table 2. Analysis of somatic metaphases showed that the chromosome number of the species is $2n = 2x = 24$. The formula is $2n = 2x = 24 = 8m + 8sm + 8st$. No satellite was observed in the chromosomes. The length of chromosomes varied from 1.79 to 6.28 μm , and the total haploid length was 42.72 μm . The average length of chromosomes was 3.56 μm . The CI varied from 19.78 to 50.00 μm . The analysis of the intrachromosomal and interchromosomal karyotype asymmetry indexes showed values of 34.52 and 39.89 for M_{CA} and CV_{CL} , respectively.

Minuartia elmalia

Mitotic metaphase chromosomes, karyogram and monoploid ideogram of *Minuartia elmalia* are given in Fig. 1B. The measurement data of these chromosomes are given in Table 3. Analysis of somatic metaphases showed that the chromosome number of the species is $2n = 2x = 32$. The formula is $2n = 2x = 32 = 12m + 16sm + 4st$. No satellite was observed in the chromosomes. The length of chromosomes varied from 2.42 to 6.32 μm , and the total haploid length was 64.42 μm . The average length of chromosomes was 4.02 μm . The CI varied from 20.52 to 45.37 μm . The analysis of the intrachromosomal and interchromosomal karyotype asymmetry indexes showed values of 29.33 and 29.35 for M_{CA} and CV_{CL} , respectively.

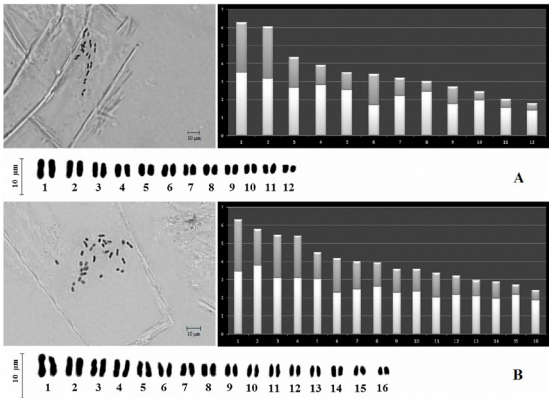


Fig. 1. Somatic metaphase chromosomes, karyograms and ideograms. A: *Minuartia mesogitana* subsp. *mesogitana*. B: *Minuartia elmalia*.

DISCUSSION

The cytogenetic characters, especially chromosome number, chromosome size and chromosome asymmetry, are useful characters in plant cytotaxonomy. They are also important to elucidate the origin, speciation and phylogenetic

relationships of plants (Stebbins, 1971). The chromosome numbers of *Minuartia mesogitana* subsp. *mesogitana* and *Minuartia elmalia* are $2n = 24$ and 32 . Both species have small chromosomes (range $1.79\text{-}6.32\ \mu\text{m}$). The chromosomes showed little variation in size within the complement (Tables 2 and 3).

The reported chromosome numbers of *Minuartia mesogitana* subsp. *mesogitana* were $2n = 22$ and 24 (Çelebioglu & Favarger, 1984, 1990; Kamari *et al.*, 1996). Çelebioglu & Favarger (1990) reported the similar results on the other subspecies. These subspecies are *Minuartia mesogitana* subsp. *brachycarpa* (Boiss. & Balansa) McNeill ($2n = 24$), *Minuartia mesogitana* subsp. *kotschyana* (Boiss.) McNeil ($2n = 22, 24$) and *Minuartia mesogitana* subsp. *lydia* (Boiss.) McNeill ($2n = 24$). According to the locality in Table 1, the diploid chromosome number of *Minuartia mesogitana* subsp. *mesogitana* is $2n = 24$. The diploid chromosome number was $2n = 22$ in different locations. Cytological data of this taxon are reported for the first time.

Aytaç & Duman (2004) reported that *Minuartia dianthifolia* Hand.-Mazz. subsp. *elmalia* is a subspecies. Parolly & Eren (2006) considered

Table 2. The measurement data of the chromosomes of *Minuartia mesogitana* subsp. *mesogitana*. TCL: total chromosome length, l: long arm, s: short arm, r: arm ratio, CI: centromeric index, m: median, sm: submedian, st: subterminal.

Chromosome pair	TCL (μm)	l (μm)	s (μm)	r	Chromosome type	CI
1.00	6.28	3.47	2.81	1.23	m	44.74
2.00	6.06	3.16	2.90	1.09	m	47.84
3.00	4.36	2.66	1.70	1.56	m	38.99
4.00	3.91	2.81	1.10	2.55	sm	28.08
5.00	3.51	2.51	1.00	2.51	sm	28.53
6.00	3.40	1.70	1.70	1.00	m	50.00
7.00	3.20	2.20	1.00	2.20	sm	31.25
8.00	3.02	2.42	0.60	4.03	st	19.78
9.00	2.71	1.73	0.98	1.77	sm	36.04
10.00	2.46	1.96	0.50	3.92	st	20.37
11.00	2.02	1.52	0.50	3.04	st	24.75
12.00	1.79	1.39	0.40	3.48	st	22.33

Table 3. The measurement data of the chromosomes of *Minuartia elmalia*. TCL: total chromosome length, l: long arm, s: short arm, r: arm ratio, CI: centromeric index, m: median, sm: submedian, st: subterminal.

Chromosome pair	TCL (µm)	l (µm)	s (µm)	r	Chromosome type	CI
1	6.32	3.46	2.86	1.21	m	45.17
2	5.78	3.77	2.01	1.88	sm	34.71
3	5.47	3.09	2.38	1.30	m	43.51
4	5.43	3.10	2.33	1.33	m	42.85
5	4.49	3.01	1.48	2.03	sm	32.96
6	4.19	2.29	1.90	1.21	m	45.37
7	4.01	2.47	1.54	1.60	m	38.40
8	3.95	2.62	1.33	1.97	sm	33.67
9	3.60	2.29	1.31	1.75	sm	36.39
10	3.60	2.34	1.26	1.86	sm	35.05
11	3.37	2.03	1.34	1.51	m	39.74
12	3.19	2.17	1.02	2.13	sm	31.99
13	2.97	2.10	0.87	2.41	sm	29.33
14	2.90	1.97	0.93	2.12	sm	32.10
15	2.73	2.17	0.56	3.88	st	20.52
16	2.42	1.85	0.57	3.25	st	23.54

that *Minuartia elmalia* is a good species. The chromosome numbers of *Minuartia elmalia* (our data) and *Minuartia dianthifolia* (Çelebioglu & Favarger, 1983) were similar.

Karyotype asymmetry is well known parameter in karyological studies (Eroğlu, 2015). The M_{CA} and CV_{CL} value are perfectly suited for the measure of intrachromosomal and interchromosomal asymmetry (Paszko, 2006; Peruzzi & Eroğlu, 2013). The M_{CA} values of *Minuartia mesogitana* subsp. *mesogitana* and *Minuartia elmalia* are 34.52 and 29.33, respectively. The CV_{CL} values of *Minuartia mesogitana* subsp. *mesogitana* and *Minuartia elmalia* are 39.89 and 29.35, respectively. A positive correlation was observed between M_{CA} and CV_{CL} ($r = 1.000$). Both M_{CA} and CV_{CL} values increase with increasing asymmetry. The location of centromere position changes in intrachromosomal karyotype asymmetry. Besides, the sizes of large and small chromosomes are quite different in interchromosomal karyotype asymmetry (Peruzzi *et al.*, 2009). According to the M_{CA} and CV_{CL}

values, *Minuartia elmalia* is more symmetrical than *Minuartia mesogitana* subsp. *mesogitana*.

In this study, the chromosome numbers, karyotypes, ideograms and karyotype asymmetry degrees of *Minuartia mesogitana* subsp. *mesogitana* and *Minuartia elmalia* were determined. The chromosome number of *Minuartia elmalia* and the chromosomal measurements of the *Minuartia mesogitana* subsp. *mesogitana* were reported here for the first time. Different chromosome numbers ($2n = 24, 32$) were found. Turkey is located at the intersection of plant geographical regions of Irano-Turanian, Mediterranean and Euro-Siberian. For this reason, diversity and genetic variations may occur among species. Chromosome number as one of the genetic variations is extremely variable ranging from low numbers to the relatively high numbers (Eroğlu & Per, 2016). It is reported that the differences found among species in chromosome numbers, karyotype formulae and asymmetry indexes may contribute to the diversification of the genus (Seijo & Fernandez, 2003). For example,

Minuartia elmalia is different from *Minuartia mesogitana* subsp. *mesogitana* morphologically and grow in high altitudes and alpine regions (Table 1). Also, *Minuartia elmalia* is different from *Minuartia mesogitana* subsp. *mesogitana* because of having: (i) higher chromosome number, (ii) higher number of median and submedian chromosomes and (iii) a more symmetric karyotype. More data are needed to understand the role of chromosome number, karyotype formula and karyotype asymmetry in the systematics and evolution of *Minuartia*.

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