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New chromosomal data, karyotype asymmetry and polyploid variations of some *Gundelia* (Asteraceae) species from Turkey

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Abstract. The genus *Gundelia* is currently represented with 18 species, of which 16 are in Turkey. In genus *Gundelia*, the chromosomal data were reported from 12 species. In the present study, it is aimed to eliminate the deficiencies in the knowledge about chromosomal data of *Gundelia* species. In Genus *Gundelia*, only a single chromosome number had been detected as $2n=18$ so far. The chromosome numbers of four species were reported here for the first time: *G. armeniaca*, *G. cappadocica*, *G. siirtica*, and *G. tehranica*. In addition, the polyploidy in the genus was rare and *G. anatolica* was identified as the first polyploid species. All karyotypes except *G. tehranica* were symmetrical, consisting of metacentric and submetacentric chromosomes. Secondary constrictions were observed in the distal regions of the long arms of the longest metacentric and submetacentric chromosomes. Thus, the chromosomal data of all Turkish *Gundelia* species were completed. In conclusion, the present study presented new data into the karyological records relating the karyotype evolution and interspecific relations of genus *Gundelia*.

Keywords: *Gundelia*, karyology, polyploidy, dysploidy, Anatolia.

INTRODUCTION

Gundelia was first collected by Leonhard Rauwolf during his travel to the Levant between 1573 and 1575 for his herbal medicine and botanical collections. Rauwolf and some have confused it with Dioscorides' *Slybum*, which today is considered to be *Silybum marianum* (L.) Gaertn. (Asteraceae). Based on Rauwolf's description and the similarity of the synflorescens of several *Eryngium* L. (Apiaceae) species, Morison recommended the name - *Eryngium Surgerycum foliis Chamaeleontis longis & spinosis* - at 17th century (Hind 2013). Linnaeus described *G. tournefortii* in 1753, after which many authors accepted *Gundelia tournefortii* monospecifically and the described taxa thereafter synonymous of it (Linnaeus 1753; Komarov 1961; Kupicha 1975; Feinbrun Dothan 1978; Meikle 1985; Rechinger 1989). The genus is currently represented with 18 species, of which 16 (12 endemic) are in Turkey (Vitek et al. 2010, 2014, 2017; Nersesyan 2014; Armağan 2016; Fırat 2016, 2017a, 2017b, 2017c; Vitek and Noroozii 2017; Vitek 2018; Vitek and Armağan 2023).

Due to the monotypic genus of *Gundelia*, not many investigations have been studied with molecular techniques. In the first studies, the phylogenetic position of the genus *Gundelia* in the family was determined (Karis

et al. 2001, Funk et al. 2004, Panero and Croizer 2016). According to Panero and Croizer (2016), the genus is a member of the subfamily Cichorioideae. After the new species started to be published, whether the species were phylogenetically supported became a matter of curiosity. Firstly, with limited species and samples, Vitek et al. (2010) and Tarıkahya-Hacıoğlu and Fırat (2017) were studied using the internal transcribed spacer marker. Finally, in 2021, an updated and expanded phylogeny based on DNA sequences of both ITS and ndhF genes was published by Ateş et al. (2021).

Gundelia genus has special inflorescence different from other genus in the Asteraceae family. The synflorescens of *Gundelia* as a whole in inflorescence is a composition of partial synflorescens (disseminules) in the axils of bracts. These partial synflorescens (disseminules) consist of 3-9 flowers and this number is an important characteristic structure for each species (Classen-Bockhoff et al. 1989).

Gundelia grows in the semi-humid to dry meadows, mountain (steppe) meadows, dry slope areas. The evidence showed that the center of diversity of *Gundelia* is Turkey, even if the genus is distributed in the area from Armenia to Egypt and from Turkey to Afghanistan (Figure 1). *Gundelia* is present everywhere in Turkey except the Black Sea, Marmara and coastal Aegean regions. The

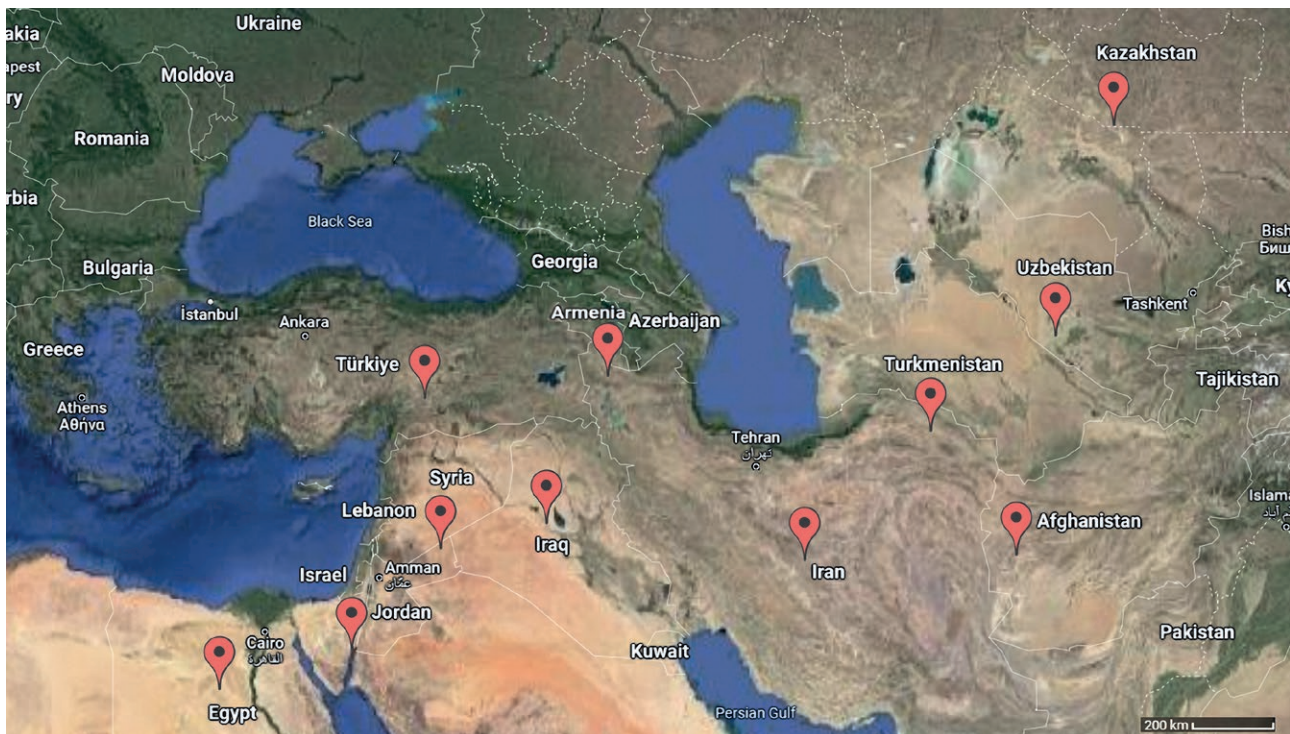


Figure 1. The distribution map of genus *Gundelia*.

geographical boundaries play a decisive role in their distribution (Karis et al. 2001; Vitek 2018).

The species belonging to the genus *Gundelia* are called “Kenger” in Turkish, “Akub” in Arabic and “Kuub” in Persian, and there are some local dialectical differences. Young shoots are used as vegetables in dishes and pickles. The latex obtained from the cut shoots is dried and used as gum. Roasted fruits are consumed like coffee. It is also used as animal food in some regions.

Cytotaxonomy, one of the sub-branches of taxonomy, uses karyological parameters in the classification of organisms. In this context, chromosomal configurations are used to understand the relationships between species and based on the assumption that “closely related species show similar chromosomal configurations”. Thus, karyotype evolution or interspecies relationships can be reconstructed by exploiting karyological similarities or variations. The chromosome number, chromosome structure and chromosomal behaviors stand out as important parameters of cytotaxonomy, and especially basic chromosome number (x), diploid chromosome number ($2n$) and karyotype asymmetry are the most preferred parameters (Eroğlu et al. 2020; Martin et al. 2020; Eroğlu et al. 2021; Kavcı et al. 2022). In genus *Gundelia*, the chromosomal data were reported from 12 species. All species were represented by only one base number ($x = 9$) and there were no reports of polyploidy (Al-Taey and Hossain 1984; Genç and Fırat 2019).

In Turkish species of *Gundelia*, the chromosomal data were reported from 12 species, which were *G. anatolica*, *G. asperrima*, *G. cilicica*, *G. colemerikensis*, *G. dersim*, *G. glabra*, *G. komagenensis*, *G. mesopotamica*, *G. munzuriensis*, *G. rosea*, *G. tournefortii*, and *G. vitekii* (Al-Taey and Hossain 1984; Genç and Fırat 2019). There was no record of the chromosome number of four species, which were *G. armeniaca*, *G. cappadocica*, *G. siirtica*, and *G. tehranica*. Due to the lack of some chromosome reports from Turkey, where there are many species of the genus, some cytotaxonomic knowledge is lacking. In this study, it is aimed to complete the missing chromosomal data in Turkish *Gundelia* species.

MATERIALS AND METHODS

Plant material

Within the scope of this study, sixteen *Gundelia* taxa distributed in different localities of Turkey were evaluated karyologically. The evaluated samples were collected from natural habitats by Dr. Metin Armağan et al. Table 1 represents the collection information and distribution regions. Turkish *Gundelia* species whose chro-

mosomal data were reported in previous studies were collected from different localities to investigate chromosomal variations.

Chromosome preparation

The plant seeds were germinated between moist Whatman papers and pretreated by α -monobromonaphthalene at 4°C for 16 h. Then, root tips were fixed by fixative solution (3 absolute alcohol and 1 glacial acetic acid - v:v) at 4°C for 24 h. The fixed root tips were stored in ethanol (70%) at 4°C. Then, root tips were hydrolyzed in 1 N hydrochloric acid at room temperature for 10 min and stained in aceto-orcein (2%). Then, squash preparations were prepared by acetic acid (45%). The preparations were frozen in liquid nitrogen, dried at room temperature, and stabilized with Depex medium (Eroğlu et al. 2020; Martin et al. 2020; Eroğlu et al. 2021; Kavcı et al. 2022).

Karyotype analysis

Well-spread ten metaphase plates were used to detect for the chromosome numbers of all species. Detailed chromosomal measurements of four species whose chromosome number was investigated for the first time were made by Karyotype software. The following parameters and formulae were used to chromosome characterizations karyotype analysis: short arm length of chromosome (p), long arm length of chromosome (q), total chromosome length (p + q), total haploid length (THL), mean haploid length (MHL), relative length (RL) = [(p + q) / THL] \times 100, and centromeric index (CI) = [p / (p + q)] \times 100. The karyotype formulae were detected based on centromere position (Levan et al. 1964) and the monoploid ideograms were drawn.

The following formulae were used to determine the intrachromosomal asymmetry (M_{CA}) and interchromosomal asymmetry (CV_{CL}): $M_{CA} = [\text{mean}(q_t - p_t) / (q_t + p_t)] \times 100$; q_t , total length of long arms and p_t , total length of short arms (Peruzzi and Eroğlu 2013). $CV_{CL} = (S_{CL} / X_{CL}) \times 100$; S_{CL} , standard deviation in a chromosome set and X_{CL} , mean chromosome length in a chromosome set (Paszko 2006).

RESULTS

Chromosome records of 16 species are herein provided (Figure 2), four of which are reported for the first time (*G. armeniaca*, *G. cappadocica*, *G. siirtica*, and *G.*

Table 1. Collection information and voucher specimens of *Gundelia* taxa.

Species (alphabetically)	Locality	Voucher
<i>G. anatolica</i> Fırat	Konya: Karapınar, S of Meke Crater Lake, 1080 m, volcanic dune (steppe), 37 40 22.3 N 33 38 32.4 E, 16 May 2016	M.Armağan 6734
<i>G. armeniaca</i> Nersesian	Muş: Karabey, 1400 m, steppe, 38 55 50.2 N 41 10 40.0 E, 01 June 2015	M.Armağan 6552
<i>G. asperrima</i> (Trautv.) Fırat	Bingöl: between Elazığ and Bingöl, Kuruca Pass, 1720 m, meadows, 38 57 22,0 N 40 14 50,6 E, 21 June 2017	M.Armağan 7544
<i>G. cappadocica</i> Fırat	Nevşehir: Avanos, Bozca, 1070 m, steppe (on calcareous soils), 38 46 06.2 N 34 59 56.9 E, 30 May 2021	O.Tugay 18.345
<i>G. cilicica</i> Fırat	Mersin: Erdemli, Tozlu, 1565 m, on degraded fields of <i>Juniperus</i> sp. forest (steppe), 36 49 14.4 N 34 08 42.1 E, 25 June 2020	M.Armağan Obs-MTN55, O.Tugay, E.Karahisar
<i>G. colemerikensis</i> Fırat	Van: Başkale, between Kovalpınar and Ömerabat, 1960 m, dry slopes, 37 49 35.8 N 44 04 20.0 E, 24 June 2020	M.Armağan 8335
<i>G. dersim</i> Vitek, Yüce & Ergin	Tunceli: Ovacık, Adaköy, 1285 m, meadows, 39 20 45.2 N 39 06 53.6 E, 29 May 2020	M.Armağan 8308, M.Özel, R.Karapınar
<i>G. glabra</i> Mill.	Elazığ: Yukarıbağ, 856 m, steppe, 38 38 01.8 N 39 34 54.6 E, 28 May 2020	M.Armağan 8306
<i>G. komagenensis</i> Fırat	Malatya: Battalgazi, NE of Beydağı, 1080 m, 38 18 10.0 N 38 27 23.6 E, 18 July 2020	M.Armağan Obs-MTN58, O.Tugay, E.Karahisar
<i>G. mesopotamica</i> Fırat	Mardin: Artuklu, Avcılar, 715 m, steppe on limestone bedrock, 37 17 53.8 N 40 42 12.9 E, 26 May 2020	M.Armağan 8293
<i>G. munzuriensis</i> Vitek, Yüce & Ergin	Tunceli: Ovacık, above Gözeler, Munzur Mountains, 2500 m, mountain steppe, 39 26 35,9 N 39 14 22,4 E, 15 July 2020	M.Armağan 8358, M.Özel, R.Karapınar, K.Es.
<i>G. rosea</i> M.Hossain & Al-Taey	Şırnak: Uludere, Güzelyazı, Hakkari-Şırnak roadside, 1380 m, dry slopes, 37 22 28.3 N 42 59 47.6 E, 23 June 2020	M.Armağan 8328, O.Tugay
<i>G. siirtica</i> Fırat	Siirt: Kurtalan, Erdurağı, mountain (steppe) meadows, 926 m, 37 53 59.1 N 41 35 32.2 E, 30 July 2020	M.Armağan 8300
<i>G. tehranica</i> Vitek & Noroozi	Şırnak: Silopi, Dedeler740 m, dry slopes, 37 19 18.5 N 42 25 32.8 E, 23 June 2020	M.Armağan 8325, O.Tugay
<i>G. tournefortii</i> L.	Karaman: Ermenek, Aşağıakın, 1110 m, steppe, 36 53 47.8 N 33 01 06.2 E, 22 August 2020	O.Tugay 17786, E.Karahisar
<i>G. vitekii</i> Armağan	Tunceli: Ovacık, Şahverdi, Mercan Valley, 1623 m, steppe, 39 27 10.8 N 39 23 51.9 E, 14 July 2020	M.Armağan 8309, M.Özel, R.Karapınar, K.Es.

tehranica), one presents polyploidy for the first time (*G. anatolica*), and twelve agree previous reports. Table 2 shows the chromosome numbers of present and previous reports. Except for polyploidy (Figure 3), the only one chromosome number detected was $2n = 18$.

Detailed chromosomal data and monoploid ideograms of the four species, whose chromosome numbers were reported for the first time were given in Table 3 and Figure 4. The smallest chromosome length among the species was 3.94 μm , in *G. siirtica*. The largest chromosome length was detected in *G. tehranica*, with 8.65 μm . The smallest total haploid length was 44.37 μm , in *G. siirtica*, and the highest value was 51.71 μm , in *G. armeniaca* and *G. cappadocia*.

Genus *Gundelia* was a monobasic genus by $x = 9$ with ploidy levels of $2x$ and $4x$. Fifteen species were diploid with $2n = 2x = 18$. *G. anatolica* was diploid and polyploid, which revealed only one polyploidy level of tetraploidy ($2n = 4x = 36$).

All species except *G. tehranica* had median (m) and submedian (sm) chromosomes, but not subtelocentric (st) and telocentric (t) chromosomes. Two different karyotype formulae were observed, which were $14m + 4sm$ and $12m + 2sm + 2st$. Secondary constrictions were observed in the distal regions of the long arms of the longest metacentric and submetacentric chromosomes (Figure 4).

In intrachromosomal asymmetry, M_{CA} value ranged from 9.98 (*G. siirtica*) to 10.63 (*G. tehranica*), which referred to symmetric karyotypes. In interchromosomal asymmetry, CV_{CL} value ranged from 13.82 (*G. armeniaca*) to 23.75 (*G. tehranica*), which referred to karyotype heterogeneity (Table 3).

DISCUSSION

Only one chromosome number excluding polyploidy was detected as $2n = 18$, which was the only diploid

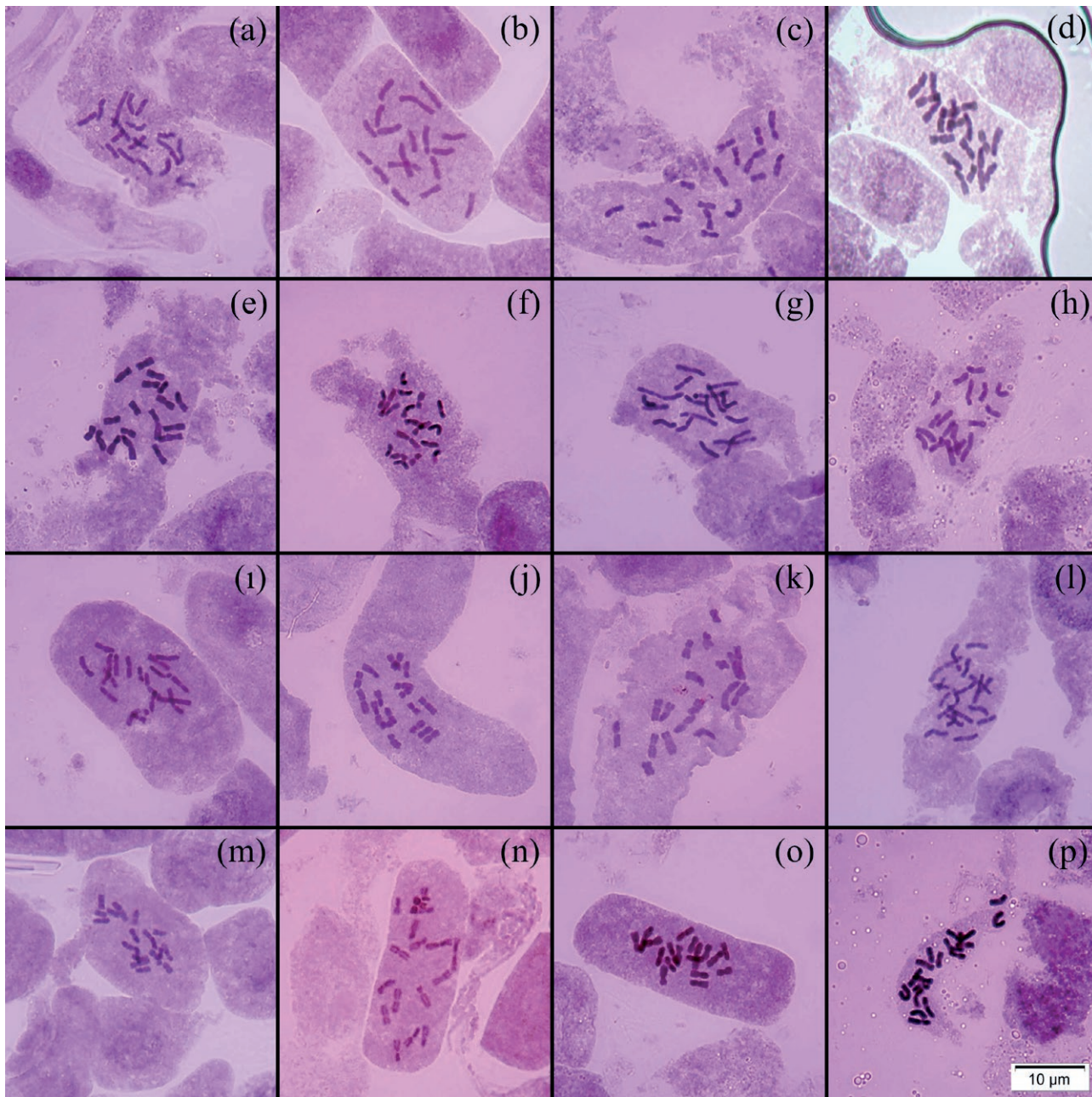


Figure 2. Somatic metaphase chromosomes of Turkish *Gundelia* species. (a) *G. armeniaca*; (b) *G. cappadocica*; (c) *G. siirtica*; (d) *G. tehranica*; (e) *G. anatolica*; (e) *G. asperrima*; (e) *G. cilicica*; (e) *G. colemerikensis*; (e) *G. dersim*; (e) *G. glabra*; (e) *G. komagenensis*; (e) *G. mesopotamica*; (e) *G. munzuriensis*; (e) *G. rosea*; (e) *G. tournefortii*; and (e) *G. vitekii*.

number ever reported in the genus. The chromosome numbers of four species were reported here for the first time: *G. armeniaca*, *G. cappadocica*, *G. siirtica*, and *G. tehranica*. The chromosome numbers were the same as in previous reports in 12 species, which were *G. anatolica*, *G. asperrima*, *G. cilicica*, *G. colemerikensis*, *G. dersim*, *G. glabra*, *G. komagenensis*, *G. mesopotamica*, *G. munzu-*

riensis, *G. rosea*, *G. tournefortii*, and *G. vitekii* (Al-Taey and Hossain 1984; Genç and Fırat 2019).

A basic chromosome number of $x = 9$ dominates in genus *Gundelia* and the genus is monobasic. The absence of basic number variations in genus *Gundelia* indicated that the mechanism of dysploidy probably did not occur in the karyotype evolution of the genus. Because

Table 2. The chromosome numbers of Turkish *Gundelia* in present and previous studies. All species were studied in this study. Turkish *Gundelia* species whose chromosomal data were reported in previous studies were collected from different localities to investigate chromosomal variations.

Species (alphabetically)	Previous results $x =$ basic number, $2n$ (ploidy level)	References	Presents results $x =$ basic number, $2n$ (ploidy level)	Observation
<i>G. anatolica</i>	$x = 9, 2n = 18$ (diploid)	Genç and Firat 2019	$x = 9, 2n = 18$ (diploid) $x = 9, 2n = 36$ (polyploid)	Equal count First report
<i>G. armeniaca</i>			$x = 9, 2n = 18$ (diploid)	First report
<i>G. asperima</i>	$x = 9, 2n = 18$ (diploid)	Genç and Firat 2019	$x = 9, 2n = 18$ (diploid)	Equal count
<i>G. cappadocica</i>			$x = 9, 2n = 18$ (diploid)	First report
<i>G. cilicica</i>	$x = 9, 2n = 18$ (diploid)	Genç and Firat 2019	$x = 9, 2n = 18$ (diploid)	Equal count
<i>G. colemerikensis</i>	$x = 9, 2n = 18$ (diploid)	Genç and Firat 2019	$x = 9, 2n = 18$ (diploid)	Equal count
<i>G. dersim</i>	$x = 9, 2n = 18$ (diploid)	Genç and Firat 2019	$x = 9, 2n = 18$ (diploid)	Equal count
<i>G. glabra</i>	$x = 9, 2n = 18$ (diploid)	Genç and Firat 2019	$x = 9, 2n = 18$ (diploid)	Equal count
<i>G. komagenensis</i>	$x = 9, 2n = 18$ (diploid)	Genç and Firat 2019	$x = 9, 2n = 18$ (diploid)	Equal count
<i>G. mesopotamica</i>	$x = 9, 2n = 18$ (diploid)	Genç and Firat 2019	$x = 9, 2n = 18$ (diploid)	Equal count
<i>G. munzuriensis</i>	$x = 9, 2n = 18$ (diploid)	Genç and Firat 2019	$x = 9, 2n = 18$ (diploid)	Equal count
<i>G. rosea</i>	$x = 9, 2n = 18$ (diploid)	Al-Taey and Hossain 1984 Genç and Firat 2019	$x = 9, 2n = 18$ (diploid)	Equal count
<i>G. siirtica</i>			$x = 9, 2n = 18$ (diploid)	First report
<i>G. tehranica</i>			$x = 9, 2n = 18$ (diploid)	First report
<i>G. tournefortii</i>	$x = 9, 2n = 18$ (diploid)	Al-Taey and Hossain 1984 Genç and Firat 2019	$x = 9, 2n = 18$ (diploid)	Equal count
<i>G. vitekii</i>	$x = 9, 2n = 18$ (diploid)	Genç and Firat 2019	$x = 9, 2n = 18$ (diploid)	Equal count

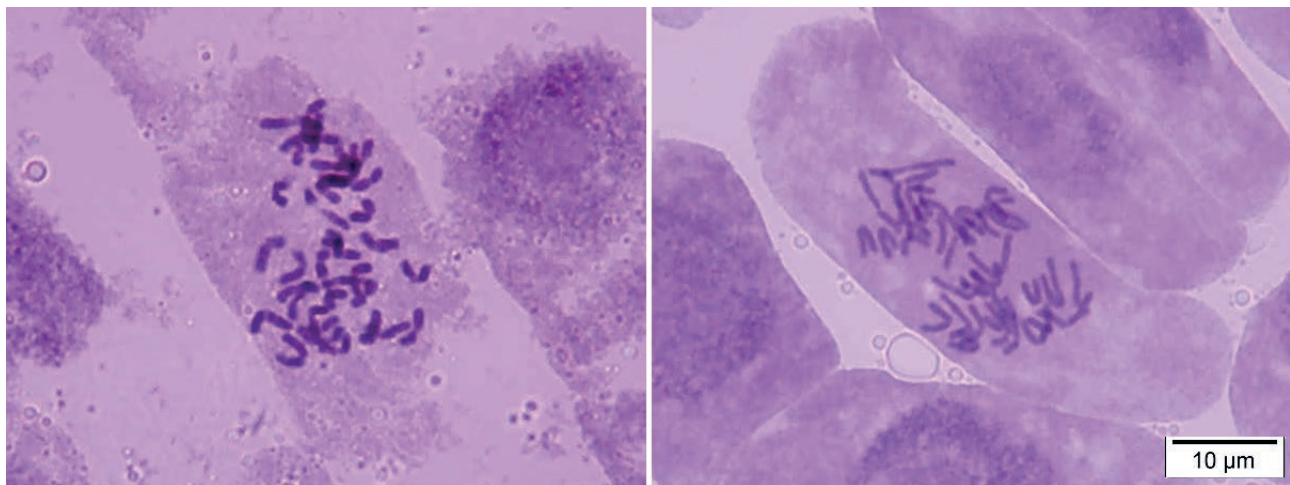


Figure 3. The polyploidy of *Gundelia anatolica*.

dysploidy causes basic number variations by fusion of metacentric chromosomes or reciprocal translocations (Eroğlu et al. 2020; Martin et al. 2022). In addition, the polyploidy in the genus was rare and *G. anatolica* was identified as the first polyploid species.

Fifteen species had metacentric and submetacentric chromosomes and only one species had subtelocentric

chromosomes, whereas no telocentric (t) chromosomes were observed. Two different karyotype samples were observed, which were m-sm and m-sm-st including secondary constrictions. Thus, five chromosome types were determined according to the positions of the primary and secondary constrictions: (i) metacentric (ii) meta-

Table 3. The detailed chromosomal data and asymmetry indices of species whose chromosome number was reported for the first time (KF: karyotype formula, SC: the shortest chromosome length, LC: the longest chromosome length, RL: relative length, CI: centromeric index, THL: total haploid length, MHL: mean haploid length, M_{CA} : mean centromeric asymmetry, CV_{CL} : coefficient of variation of chromosome length).

	<i>G. armeniaca</i>	<i>G. cappadocica</i>	<i>G. siirtica</i>	<i>G. tehranica</i>
KF	14m + 4sm	14m + 4sm	14m + 4sm	12m + 2sm + 2st
SC (µm)	4.80	4.57	3.94	4.01
LC (µm)	7.10	8.51	6.97	8.65
RL (%) SC–LC	9.28–13.73	8.84–16.46	8.88–15.71	7.92–17.08
CI (min–max)	33.82–49.45	25.46–49.30	32.87–49.43	23.02–49.80
THL	51.71	51.71	44.37	50.65
MHL	5.75	5.75	4.93	5.63
M_{CA}	10.02	10.53	9.98	10.63
CV_{CL}	13.82	22.72	21.30	23.75

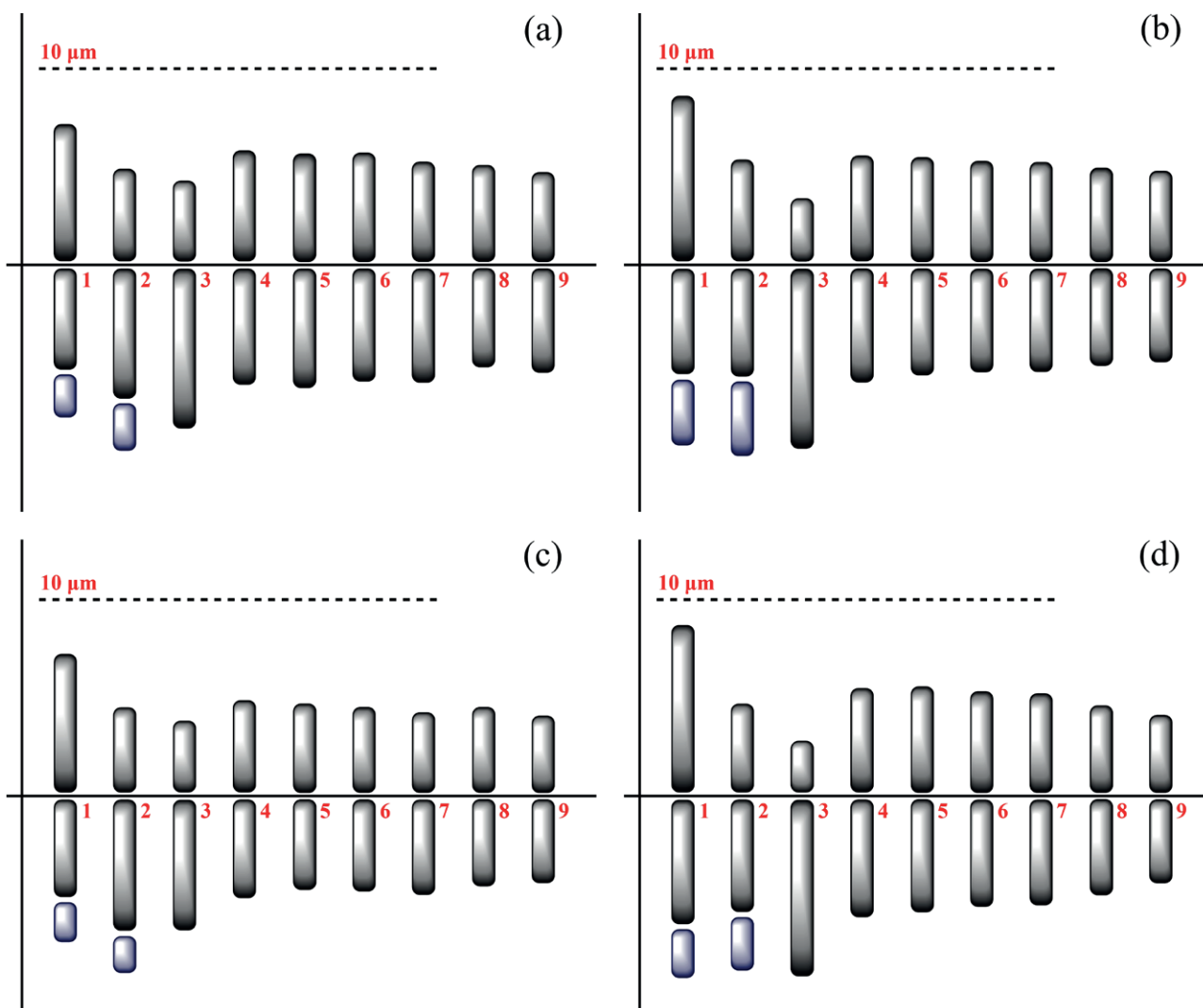


Figure 4. The monoploid ideograms of the species whose chromosome number was reported for the first time. (a) *G. armeniaca*; (b) *G. cappadocica*; (c) *G. siirtica*; (d) *G. tehranica*.

of the long arm, (iii) submetacentric, (iv) submetacentric with secondary constriction in the distal region of the long arm, (v) subtelocentric. In twelve *Gundelia* species, Genç and Fırat (2019) reported that the secondary constrictions at short or long arms of submetacentric chromosomes and in the distal region of long arm of the longest metacentric chromosome.

In intrachromosomal asymmetry, all karyotypes were symmetric. The most symmetric and asymmetrical karyotypes were the karyotypes of *G. siirtica* and *G. tehranica*, respectively. In interchromosomal asymmetry, all karyotypes were symmetric. The most symmetric and asymmetrical karyotypes were the karyotypes of *G. armeniaca* and *G. tehranica*, respectively. Genç and Fırat (2019) reported that *G. rosea* and *G. tournefortii* had the relatively high intrachromosomal asymmetry and low intrachromosomal asymmetry, respectively; also, *G. vitekii* and *G. anatolica* had the high interchromosomal and low interchromosomal asymmetry, respectively.

In the present study, it was recorded only one chromosome number ($2n = 18$) excluding polyploidy ($2n = 36$), the first report for diploid numbers of four species, the first report of polyploidy for the genus, and the same chromosome count with previous report in the twelve species. Thus, the chromosomal data of all Turkish *Gundelia* species were completed. In conclusion, the present study presented new data into the karyological records relating the karyotype evolution and interspecies relations of genus *Gundelia*. In addition, the dysploidy and polyploidy mechanisms probably did not have an important role in the speciation of genus.

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