Karyological Analysis of Endemic Five *Muscari* Taxa

Meryem Bozkurt*

Science Faculty, Department of Biology, Selçuk University, Konya, Turkey

Received July 6, 2020; accepted July 20, 2020

Summary Five endemic *Muscari* taxa in Turkey are examined in terms of chromosome number and morphology. *Muscari sivrihisardaghlarensis* ($2n=18$), *M. serpentinicum* ($2n=18$), *M. atillae* ($2n=18$) and *M. ufukii* ($2n=18$) are diploid. *M. tuzgoluense* is polyploid with $2n=36$ chromosome number. Chromosome counts have confirmed the previous reports that *Muscari* genus is $x=9$ of basic chromosome number. The chromosome number and morphology of these species are reported for the first time. The karyotypes have the predominance of metacentric and submetacentric chromosomes. Six quantitative asymmetric indices were used to evaluate the karyological characteristics of the species, and these results increase our karyological knowledge about the species.

Keywords Chromosome number, Endemic, Karyomorphology, Karyotype, *Muscari*, Turkey.

The genus *Muscari* Miller, which is distributed in Europe, the Mediterranean region, and Southwest Asia, is represented by about 54 species (Jafari and Maassoumi 2011). Some species of the genus are used as ornamental plants in the park and gardens (Ferrauto and Pavone 2016). In Turkey, *Muscari* is represented by 44 species, of which 31 endemics (Demirci Kayıran *et al.* 2019, Doğu and Uysal 2019, Eker 2019a, b, Eker *et al.* 2019, Yıldırım and Kılıç 2019). When considering the density of the species number and endemism rate in *Muscari*, Turkey is one of the main differentiation and gene centers (Yıldırım 2016, Böhnert and Lobin 2017, Doğu and Uysal 2019). In *Muscari*, both the taxonomic and the karyological situations are quite complex (Davis and Stuart 1980, 1984, Speta 1989, 1998, Karlén 1991, Azizi *et al.* 2016). The use of karyological data in the taxonomy can be handled as taxonomic evidence for biosystematics classification at the level of family, genus, and species and even it can contribute to evaluating the genetic relationship among populations belonging to species (Stace 1980, Odah and Oran 2007, Guerra 2008). The main goal of this study is to reveal the karyomorphological features of Turkish endemic *Muscari* species and trying to explain the effect of chromosomal behaviors on species-related relationships.

Materials and methods

Plant materials belonging to *Muscari* five taxa were collected from natural areas of Turkey (Table 1). At least three bulbs each taxon studied were germinated in suitably prepared foams in the water. Chromosome

count was performed by the crushing technique (Goldblatt 1996). The root tips from the bulbs were treated with 2 mM 8-hydroxyquinoline at 4°C for 8 h. After the pretreatment, the materials were fixed with Carnoy fixative at low temperature for 24 h. For the staining process, the materials were hydrolyzed with 5 M HCl for 1 h at room temperature and they were stained with 1% aceto-orcin. After obtaining the best metaphase, the pictures of the chromosomes were taken with an Olympus DP72 digital camera mounted on an Olympus BX53 microscope. Karyotype measurements were performed with the KAMERAM program (Argenit KAMERAM GEN3) on the best metaphase images obtained and karyomorphology was determined using various symmetry indices. The morphology of chromosomes was named according to Levan *et al.* (1964). The intrachromosomal asymmetry index (A_1) and the interchromosomal asymmetry index (A_2) proposed by Romero Zarco (1986) were used. Karyotype asymmetry was estimated using the coefficient of variation of chromosome length (CV_{CL}), the coefficient of variation of the centromeric index (CV_{CI}), and karyotype asymmetry index (AI) proposed by Paszko (2006). The mean centromeric asymmetry (M_{CA}) was also calculated according to the formula given by Peruzzi and Eroğlu (2013). The karyograms of taxa were made using the KAMERAM program.

Results

In this study, chromosome number and morphology of five *Muscari* taxa are given for the first time (Tables 2, 3).

M. sivrihisardaghlarensis, which is distributed in Central Anatolia, is a rare endemic species and it has a diploid chromosome number of $2n=18$ (Figs. 1A, 2A).

* Corresponding author, e-mail: mbozkurt@selcuk.edu.tr
DOI: 10.1508/cytologia.85.289

Table 1. Taxa studied and localities.

Subgenus	Taxon	Locality
<i>Botryanthus</i>	<i>M. sivrihisardaghlarens</i>	Sivrihisar-Karacaören, Eryiğit radar station, <i>J. excelsa</i> - <i>J. oxycedrus</i> clearings, 1,662 m, 07 iv 2018, <i>H. Demirelma</i> 3360 and <i>H. Dural</i> .
<i>Botryanthus</i>	<i>M. serpentanicum</i>	Muğla, Çamobası, 1,444 m, 02 iv 2018, <i>T. Uysal</i> 3442 and <i>M. Bozkurt</i> .
<i>Botryanthus</i>	<i>M. atillae</i>	Malatya, Akçadag, Levent canyon, southeast slopes of Çayözü village, 1,110 m, 28 iv 2018, <i>T. Uysal</i> 3618.
<i>Botryanthus</i>	<i>M. tuzgoluense</i>	Konya, Gölyazı-Eskil road 2 km, halophytic steppe, 950 m, 02 v 2018, <i>T. Uysal</i> 3639.
<i>Leopoldia</i>	<i>M. ufukii</i>	Gaziantep-Osmaniye road, 38 km from Gaziantep, oak- <i>Pistacia</i> formation clearings to the right of the road, 1,096–1,115 m, 04 vi 2018, <i>T. Uysal</i> 3728 and <i>M. Bozkurt</i> .

Table 2. The chromosome features of *Muscari* taxa.

Taxa	2n	SC-LC (μ m)	LC/SC	p (μ m) ($\pm$ SD)	q (μ m) ($\pm$ SD)	CL (μ m) ($\pm$ SD)	TCL (μ m)	CI ($\pm$ SD)	KF
<i>M. sivrihisardaghlarens</i>	18	1.22–2.31	1.903	0.72 ($\pm$ 0.09)	0.93 ($\pm$ 0.27)	1.65 ($\pm$ 0.35)	14.846	44 ($\pm$ 0.04)	16m+2sm
<i>M. serpentanicum</i>	18	1.15–3.22	2.797	0.70 ($\pm$ 0.14)	1.13 ($\pm$ 0.64)	1.83 ($\pm$ 0.73)	16.469	42 ($\pm$ 0.09)	14m+2sm+2st
<i>M. atillae</i>	18	1.13–3.14	2.771	0.73 ($\pm$ 0.19)	1.17 ($\pm$ 0.52)	1.89 ($\pm$ 0.69)	17.013	40 ($\pm$ 0.06)	12m+6sm
<i>M. tuzgoluense</i>	36	0.99–1.83	1.856	0.61 ($\pm$ 0.11)	0.80 ($\pm$ 0.19)	1.41 ($\pm$ 0.29)	25.44	44 ($\pm$ 0.03)	32m+4sm
<i>M. ufukii</i>	18	1.58–4.77	3.01	1.01 ($\pm$ 0.24)	1.85 ($\pm$ 0.94)	2.86 ($\pm$ 1.14)	25.729	38 ($\pm$ 0.08)	10m+6sm+2st

SC: Shortest chromosome length, LC: longest chromosome length, p: mean length of the long and short arm, q: mean length of the long arm, CL: mean chromosome length, TCL: total haploid chromosome length, CI: mean centromeric index, SD: standard deviation, KF: karyotype formula, m: metacentric, sm: submetacentric, st: subtelocentric.

Table 3. The karyotype indices of *Muscari* taxa.

Taxa	A ₁	A ₂	CV _{CL}	CV _{CI}	AI	M _{CA}
<i>M. sivrihisardaghlarens</i>	0.191	0.213	21.258	9.913	2.107	12.72
<i>M. serpentanicum</i>	0.258	0.398	39.826	20.552	8.185	23.49
<i>M. atillae</i>	0.325	0.366	36.609	14.349	5.253	23.15
<i>M. tuzgoluense</i>	0.215	0.205	20.517	7.722	1.584	13.47
<i>M. ufukii</i>	0.356	0.399	39.896	20.421	8.147	29.37

Fig. 1. Mitotic metaphase chromosomes of *Muscari* taxa. A: *M. sivrihisardaghlarens*, B: *M. serpentanicum*, C: *M. atillae*, D: *M. tuzgoluense*, E: *M. ufukii*.

The chromosome number and morphology of this species was determined for the first time. The karyotype formula (KF: 16m+2sm), coefficient of variation of chromosome length (CV_{CL}: 21.258), and coefficient of variation of the centromeric index (CV_{CI}: 9.913) of this species were given in Tables 1 and 2.

M. serpentanicum, which is known from a single locality in the Sandras mountain in Muğla (southwestern Anatolia region of Turkey), is a local endemic species and it has a diploid chromosome number of 2n=18 (Figs. 1B, 2B). The chromosome number and morphology of this species was determined for the first time. KF

Fig. 2. Karyograms of *Muscari* taxa. A: *M. sivrihisardaghlarensis*, B: *M. serpentinicum*, C: *M. atillae*, D: *M. tuzgoluense*, E: *M. ufukii*.

(14m+2sm+2st), CV_{CL} (39.826), and CV_{CI} (20.552) of this species were given in Tables 1 and 2.

M. atillae, which is known from a restricted area in Levent Canyon in Malatya (eastern Anatolia region of Turkey), is an endemic species and it has a diploid chromosome number of $2n=18$ (Figs. 1C, 2C). The satellite was detected in the first chromosome pair of this species. The chromosome number and morphology of this species was determined for the first time. KF (12m+6sm), CV_{CL} (36.609), and CV_{CI} (14.349) of this species were given in Tables 1 and 2.

M. tuzgoluense grown in extreme environmental conditions is an endemic species and it has a polyploid chromosome number of $2n=36$ (Figs. 1D, 2D). The chromosome number and morphology of this species was determined for the first time. KF (32m+4sm), CV_{CL} (20.517) and CV_{CI} (7.722) of this species were given in Tables 1 and 2.

M. ufukii is an endemic species and it has a diploid chromosome number of $2n=18$ (Figs. 1E, 2E). The chromosome number and morphology of this species was determined for the first time. KF (10m+6sm+2st), CV_{CL} (39.896), and CV_{CI} (20.421) of this species were given in Tables 1 and 2.

Discussion

Karyological studies for the *Muscari* have shown that the main evolutionary mechanisms involving chromosome rearrangement and polyploidy are effective in the diversification of the genus (Bentzer 1969, 1972, 1974, Bentzer and Ellmer 1975, Oliver and Ruiz-Rejón 1980, Ruiz-Rejón *et al.* 1985, Valdés and Díaz-Lifante 1992, Cuñado *et al.* 2000, De la Herrán *et al.* 2001). Therefore, the last taxonomic arrangements of the *Muscari* are based on karyological data (Garbari 1968, 1969, Demirci *et al.* 2013, Doğu and Uysal 2019, Eroğlu and Pınar 2019). Also, the recently described new species were determined the karyological features (Demirci *et al.* 2013,

Demirci Kayıran *et al.* 2019, Doğu and Uysal 2019, Eroğlu and Pınar 2019).

In this study, five species are diploid ($2n=18$) except for *M. tuzgoluense*, which is tetraploid ($2n=36$). The karyotypes of *M. sivrihisardaghlarensis*, *M. serpentinicum*, *M. atillae*, *M. tuzgoluense* and *M. ufukii* are different from each other (16m+2sm, 14m+2sm+2st, 12m+6sm, 32m+4sm, and 10m+6sm+2st, respectively) and mostly consist of metacentric chromosomes.

Yıldırım and Selvi (2002) reported that *M. sivrihisardaghlarensis* is morphologically closely related to the species *M. armeniacum* and *M. neglectum*, but differ in terms of characteristics such as shorter and reddish scape, linear leaves, and shorter raceme. Besides the morphological differences, karyotypes of this endemic species (Table 2) are quite distinct from *M. armeniacum* and *M. neglectum* reported (Table 4; Karlén 1984, Jafari *et al.* 2008, Demirci Kayıran and Özhatay 2017).

Pirhan *et al.* (2014) reported that *M. serpentinicum* related to *M. commutatum* and *M. latifolium*. In previous karyotypes of *M. commutatum* and *M. latifolium*; in the idiogram of *M. commutatum*, Karlén (1984) determined that the five long m to sm pairs decrease more or less continuously in length from the first to the fifth and, the four short pairs are m. He determined that the satellites are situated on the short arms of the second pair. Stuart (1970) found that *M. latifolium* has its satellites on the long arms of the second pair. Moreover, Stuart (1966) reported that the karyotypes of *M. latifolium* and *M. bourgaei* are also very much like the *Leopoldia* (Parl.) Rouy. In the karyotype of the *M. serpentinicum*, there are metacentric and submetacentric chromosomes as well as subtelocentric chromosomes. In addition, there is no satellite in the chromosomes of the *M. serpentinicum*. However, the karyotypes of *M. commutatum* and *M. latifolium* are placed on satellites (Stuart 1970, Karlén 1984).

Yıldırım (2015) reported that *M. atillae* has similarities with *M. discolor* and *M. anatolicum* in terms of

Table 4. Comparison of the results with the previously reported karyotype formulas for *Muscari* taxa.

Taxa	Karyotype formula	References
<i>M. sivrihisardaghlarensis</i>	16m+2sm (diploid)	in this study
<i>M. neglectum</i>	4m+10sm+4st ^{SAT} (diploid)	Demirci Kayıran and Özhatay (2017)
	16m+12sm+8st (tetraploid)	Demirci Kayıran and Özhatay (2017)
	35m+m ^{SAT} +18sm, 36m+18sm (hexaploid)	Demirci Kayıran and Özhatay (2017)
	n=4m+5sm (diploid, tetraploid, hexaploid, oktaploid)	Karlén 1984
	13m+11sm+2st+t, 23m+3sm, 21m+5sm, 16m+9sm+t or 16m+7sm, 13m+9sm+st, 17m+5sm+st (aneuploid)	Jafari <i>et al.</i> (2008)
	17m+5sm+st (autopentaploid)	Jafari <i>et al.</i> (2008)
	14m+3sm (tetraploid)	Jafari <i>et al.</i> (2008)
<i>M. armeniacum</i>	12m+4sm+2st ^{SAT} , 12m+4sm+2st n=7m+2sm	Demirci Kayıran and Özhatay (2017) Karlén 1984

some morphological features. Arslan and Uysal (2009) reported that there are five m and four sm pairs in the karyotype of the *M. discolor*. They also reported that satellite was placed in the long arm of the 6th chromosome pair of this species. Demirci Kayıran and Özhatay (2017) reported the karyotype of *M. anatolicum* as $2n=3x=27=15m+9sm+3st$ (triploid). We can say that *M. atillae* differs from *M. discolor* and *M. anatolicum* in terms of chromosome morphology as well as general morphological aspect.

Demirci *et al.* (2014) reported that *M. ufukii* is close to *M. longipes* and *M. tenuiflorum*. Jafari *et al.* (2008) determined the karyotype formula of *M. longipes* as $2m+4sm+3st$. The previous karyotypes of *M. tenuiflorum* were reported as $2n=18=8m+6sm+4st$ (Demirci Kayıran and Özhatay 2017), $5m+3sm+1st$ or $1m+5sm+6st+1t$ (Jafari *et al.* 2008), and $2st+2sm+14m$ (Doğu and Uysal 2019). The TCL of the *M. ufukii*, which is a member of the *Leopoldia*, is larger than the *Botryanthus* taxa (Table 2). These findings are consistent with previous reports (Stuart 1970, Jafari *et al.* 2008). According to Lima De Faria (1980), chromosomes are uniform in size; in all studied *Muscari* taxa having small chromosomes (CL: 1.41–2.91 μ m). The chromosome lengths for some taxa of the *Muscari* (CL: 1.08–5.78 μ m) are notified as small and medium-size and these sizes (Demirci Kayıran and Özhatay 2017) are largely consistent with our results. The TCL of the *M. sivrihisardaghlarensis*, *M. serpentinicum*, *M. atillae*, *M. tuzgoluense* is compatible with the values (16–37 μ m) previously reported in the *Botryanthus* (Kunth) Rouy (Demirci Kayıran and Özhatay 2017).

Polyploidy in plants has been identified as one of the main processes that cause genetic diversity and speciation (Stebbins 1950, Grant 1971, Rieseberg 1997, Wendel 2000, Doyle *et al.* 2003). Garbari (1972) claimed that the *Bellevalia* with $x=4$ basic chromosome number had the potential ancestor of the *Muscari* as a result of its polyploidy and aneuploidy. In *Muscari*, polyploidy is more common in *Botryanthus* (such as *M. parviflorum* $2n=36$; Rossi and Capineri 1982, Speta 1982, Garbari 1984, Demirci Kayıran and Özhatay 2017). In addition, many species in this subgenus (such as *M. neglectum*

species) have been reported to have ploidy levels (Davis and Stuart 1984, Karlén 1984, Valdés and Mejías 1988, Karlén 1991, Valdés 1996, Garbari 2003). A new polyploid species, *M. tuzgoluense*, for the *Botryanthus* was reported for the first time in this study.

According to Stebbins (1971), an increase in intra-chromosomal asymmetry occurs due to the change in the centromeric position of the chromosomes and symmetrical karyotype composed of m and sm is primitive. We can say that *M. sivrihisardaghlarensis* and *M. tuzgoluense*, which have the most symmetrical karyotypes, are primitive among five species studied. As a result of the analyses, the change in the centromeric position of the chromosomes and high CV_{CI} showed that the chromosomal heterogeneity of *M. serpentinicum* and *M. ufukii* is high. Another parameter used to measure intrachromosomal asymmetry is M_{CA} . The presence of symmetric and asymmetric karyotypes implies chromosomal rearrangements and different evolutionary steps (Uysal *et al.* 2017). Among the studied species, *M. ufukii* (CV_{CL} : 39.896, AI: 8.147, M_{CA} : 29.37) has the most asymmetric karyotypes and more advanced in terms of phylogeny and evolutionary process according to Stebbins (1971).

Acknowledgements

I would like to thanks a lot the foundation Tubitak (project number 117Z222) because of its financial support. I thanks Prof. Dr. Tuna UYSAL, Prof. Dr. Huseyin DURAL and Asst. Prof. Dr. Hakki DEMIRELMA for plant collecting and species identification and I would also like to thanks Prof. Dr. Tuna UYSAL for many valuable suggestions that greatly improved the manuscript.

References

- Arslan, E. and Uysal, T. 2009. A Karyological study on Endemic *Muscari discolor* Boiss & Hausskn Species. Selçuk Üniversitesi Fen Edebiyat Fakültesi Fen Dergisi **34**: 63–68. (in Turkish)
- Azizi, N., Amirouche, R. and Amirouche, N. 2016. Karyological investigations and new chromosome number reports in *Bellevalia* Lapeyrouse, 1808 and *Muscari* Miller, 1758 (Asparagaceae)

- from Algeria. *Comp. Cytogenet.* **10**: 171–187.
- Bentzer, B. 1969. Chromosome morphology in Aegean populations of *Leopoldia* Parl. (Liliaceae). *Bot. Not.* **122**: 457–480.
- Bentzer, B. 1972. Variation in the chromosome complement of *Leopoldia comosa* (L.) Parl. (Liliaceae) in the Aegean (Greece). *Bot. Not.* **125**: 406–418.
- Bentzer, B. 1974. Karyotypes and meiosis in *Leopoldia* Parl (Liliaceae) from the southern and central Aegean (Greece). *Bot. Not.* **127**: 69–86.
- Bentzer, B. and Ellmer, M. 1975. A case of stable chromosomal polymorphism in *Leopoldia comosa* (Liliaceae). *Hereditas* **81**: 127–132.
- Böhnert, T. and Lobin, W. 2017. *Leopoldia neumannii* sp. nov. (Asparagaceae, Scilloideae): A new species of *Muscari sensu lato* from Greece. *Willdenowia* **47**: 179–185.
- Cuñado, N., De la Herrán, R., Santos, J. L., Ruiz-Rejón, C., Garrido-Ramos, M. A. and Ruiz-Rejón, M. 2000. The evolution of the ribosomal loci in the subgenus *Leopoldia* of the genus *Muscari* (Hyacinthaceae). *Plant Syst. Evol.* **221**: 245–252.
- Davis, P. H. and Stuart, D. C. 1980. *Muscari* Mill. In: Tutin, T. G., Heywood, V. H. and Valentine, D. H. (eds.). *Flora Europaea* Vol. 5. Cambridge University Press, London. pp. 46–49.
- Davis, P. H. and Stuart, D. C. 1984. *Muscari* Mill. In: Davis, P. H. (ed.). *Flora of Turkey and the East Aegean Islands* 8. Edinburgh University Press, Edinburgh. pp. 245–263.
- De la Herrán, R., Robles, F., Cuñado, N., Santos, J. L., Ruiz-Rejón, M., Garrido-Ramos, M. A. and Ruiz-Rejón, C. 2001. A heterochromatic satellite DNA is highly amplified in a single chromosome of *Muscari* (Hyacinthaceae). *Chromosoma* **110**: 197–202.
- Demirci Kayıran, S. and Özhatay, F. N. 2017. A karyomorphological study on the genus *Muscari* Mill. growing in Kahramanmaraş (Turkey). *Turk. J. Bot.* **41**: 289–298.
- Demirci Kayıran, S., Özhatay, N. and Kaya, E. 2019. *Muscari tauricum* (Asparagaceae, Scilloideae), a new species from Turkey. *Phytotaxa* **399**: 109–118.
- Demirci, S., Özhatay, N., Gürdal, B. and Kaya, E. 2014. Contributions to the Turkish Geophyte Flora. In: Kaya, E. (ed.). *Muscari* Mill. Geophytes of Turkey 3. Atatürk Central Horticultural Research Institute, Edition No: 96, Yalova, pp. 543–550. (in Turkish)
- Demirci, S., Özhatay, N. and Koçyiğit, M. 2013. *Muscari erdalii* (Asparagaceae, Scilloideae), a new species from Southern Turkey. *Phytotaxa* **154**: 38–46.
- Doğu, S. and Uysal, T. 2019. *Muscari savranii* (Asparagaceae), a new species from central Anatolia, Turkey. *Phytotaxa* **402**: 155–164.
- Doyle, J. J., Doyle, J. L., Rauscher, J. T. and Brown, A. H. D. 2003. Diploid and polyploid reticulate evolution throughout the history of the perennial soybeans (*Glycine* subgenus *Glycine*). *New Phytol.* **161**: 121–132.
- Eker, İ. 2019a. *Muscari fatmacereniae* (Asparagaceae, Scilloideae), a new species from southern Anatolia. *Phytotaxa* **397**: 99–106.
- Eker, İ. 2019b. *Muscari pamiryigidii* (Asparagaceae, Scilloideae), a new species from northwestern Anatolia. *Phytotaxa* **408**: 255–266.
- Eker, İ., Yıldırım, H. and Armağan, M. 2019. A new grape hyacinth record for the flora of Turkey: *Muscari pallens* (M. Bieb.) Fisch. (Asparagaceae). *Bağbahçe Bilim Dergisi* **6**: 45–53. (in Turkish)
- Eroğlu, H. and Pinar, S. M. 2019. The taxonomic resurrection of *Muscari haradjianii* (Asparagaceae, Scilloideae), and a new synonym in the genus *Muscari* in Turkey. *Phytotaxa* **418**: 97–106.
- Ferrauto, G. and Pavone, P. 2016. Pollen morphology and taxonomic implications of *Muscari* Miller (Hyacinthaceae) species from Sicily. *Plant Biosyst.* **150**: 27–34.
- Garbari, F. 1968. The *Muscari* genus (Liliaceae): Contribution to the cytotaxonomic review. *Giorn. Bot. Ital.* **102**: 87–105. (in Italian)
- Garbari, F. 1969. New cytological observations on the *Muscari* and *Leopoldia* genera. *Giorn. Bot. Ital.* **103**: 1–9. (in Italian)
- Garbari, F. 1972. Notes on the genus *Pseudomuscari* (Liliaceae). *Webbia* **27**: 369–381. (in Italian)
- Garbari, F. 1984. Some karyological and taxonomic remarks on the Italian *Muscari* (Liliaceae). *Webbia* **38**: 139–164.
- Garbari, F. 2003. *Muscari neglectum* Guss. and *M. atlanticum* Boiss. et Reuter (Hyacinthaceae). Types, characters and comments on the two species. *Inform. Bot. Ital.* **35**: 329–336. (in Italian)
- Goldblatt, P. 1996. Index to Plant Chromosome Numbers 1992–1993. Monographs in Systematic Botany from the Missouri Botanical Garden **58**: 1–276.
- Grant, V. 1971. *Plant Speciation*. Columbia University Press, New York.
- Guerra, M. 2008. Chromosome numbers in plant cytotaxonomy: Concepts and implications. *Cytogenet. Genome Res.* **120**: 339–350.
- Jafari, A. and Maassoumi, A. A. 2011. Synopsis of *Leopoldia*, *Muscari* and *Pseudomuscari* (Hyacinthaceae) in Iran, with *Leopoldia ghoushtchiensis* sp. nova. *Ann. Bot. Fenn.* **48**: 396–400.
- Jafari, A., Maassoumi, A. A. and Farsi, M. 2008. Karyological study on *Bellevallia* and *Muscari* (Liliaceae) species of Iran. *Asian J. Plant Sci.* **7**: 50–59.
- Karlén, T. 1984. Karyotypes and chromosome numbers of five species of *Muscari* (Liliaceae). *Willdenowia* **14**: 313–320.
- Karlén, T. 1991. *Muscari* Miller. In: Strid, A. and Tan, K. (eds.). *Mountain Flora of Greece* Vol. 2. Edinburgh University Press, Edinburgh. pp. 697–701.
- Levan, A., Fredga, K. and Sandberg, A. A. 1964. Nomenclature for centromeric position on chromosomes. *Hereditas* **52**: 201–220.
- Lima De Faria, A. 1980. Classification of genes, rearrangements and chromosomes according to the chromosome field. *Hereditas* **93**: 1–46.
- Odah, M. and Oran, S. 2007. Karyological study of the genus *Ornithogalum* (Liliaceae) in Jordan, dirasat. *Pure Sci.* **34**: 210–220.
- Oliver, J. L. and Ruiz-Rejón, M. 1980. The relation between isozymes and ploidy level. Its application to biogeographical studies of *Muscari atlanticum* (Liliaceae). *Taxon* **29**: 27–32.
- Paszko, B. 2006. A critical review and a new proposal of karyotype asymmetry indices. *Plant Syst. Evol.* **258**: 39–48.
- Peruzzi, L. and Eroğlu, H. E. 2013. Karyotype asymmetry: Again, how to measure and what to measure? *Comp. Cytogenet.* **7**: 1–9.
- Pirhan, A. F., Yıldırım, H. and Altoğlu, Y. 2014. *Muscari serpentinicum* sp. nova (Asparagaceae): A new species from western Anatolia, Turkey. *Ot Sist. Bot. Dergisi* **21**: 1–14.
- Rieseberg, L. H. 1997. Hybrid origins of plant species. *Annu. Rev. Ecol. Syst.* **28**: 359–389.
- Romero-Zarco, C. 1986. A new method for estimating karyotype asymmetry. *Taxon* **35**: 526–530.
- Rossi, W. and Capineri, R. 1982. Observations on *Muscari parviflorum* Desf. in Italy. *Ann. Bot.* **40**: 107–110. (in Italian)
- Ruiz-Rejón, M., Pascual, L., Ruiz-Rejón, C., Valdés, B. and Oliver, J. L. 1985. A new species of *Muscari* subgenus *Leopoldia* from the Iberian peninsula. *Biochem. Syst. Ecol.* **13**: 239–250.
- Stace, C. A. 1980. *Plant Taxonomy and Biosystematics*. Edward Arnold, London.
- Speta, F. 1982. Über die Abgrenzung und Gliederung der Gattung *Muscari* und über ihre Beziehungen zu anderen Vertretern der Hyacinthaceae. *Bot. Jahrb Syst.* **103**: 247–291.
- Speta, F. 1989. *Muscari* (subg. *Leopoldia*) *mirum* Speta, spec. nova, im kreise seiner nächsten verwandten. *Phyton (Horn)* **29**: 105–114.
- Speta, F. 1998. Hyacinthaceae. In: Kubitzki, K. (ed.). *The Families and Genera of Vascular Plants*, 3. Monocotyledons. Springer Verlag, Heidelberg. pp. 261–285.
- Stebbins, G. L. 1950. *Variation and Evolution in Plants*. Columbia University Press, New York.
- Stebbins, G. L. 1971. *Chromosomal Evolution in Higher Plants*. Edward Arnold, London.

- Stuart, D. C. 1966. Cytotaxonomic studies in *Muscari sensu lato*. Thesis University of Edinburgh, Edinburgh.
- Stuart, D. C. 1970. Chromosome numbers in the genus *Muscari*. Notes R. Bot. Gard. Edinb. **30**: 189–196.
- Uysal, T., Bozkurt, M., Tugay, O., Ertuğrul, K., Şimşek Sezer, E. N. and Köse, Y. B. 2017. Karyomorphology of Turkish species in *Centaurea* sections *Centaurea* and *Phalolepis* (Asteraceae) and implications for taxonomy. Plant Biosyst. **151**: 949–964.
- Valdés, B. 1996. Notas sobre Liliáceas del N de Marruecos. Lagasalia **18**: 324–330.
- Valdés, B. and Díaz-Lifante, Z. 1992. Karyology and reproductive biology of *Muscari matritensis* M. Ruíz Rejón & al. (Liliaceae). Bot. J. Linn. Soc. **109**: 125–130.
- Valdés, B. and Mejías, J. A. 1988. Contribución al estudio de la biología de la reproducción de las especies españolas de *Muscari* (Liliaceae). Lagasalia **15**: 95–103.
- Wendel, J. F. 2000. Genome evolution in polyploids. Plant Mol. Biol. **42**: 225–249.
- Yıldırım, H. 2015. *Muscari atillae* sp. nova (Asparagaceae): A new species from eastern Anatolia, Turkey. Phytotaxa **213**: 291–295.
- Yıldırım, H. 2016. *Muscari elmasii* sp. nova (Asparagaceae): A new species from western Anatolia, Turkey. Turk. J. Bot. **40**: 380–387.
- Yıldırım, Ş. and Kılıç, Ö. 2019. A new species of *Muscari* Mill. (Asparagaceae/Liliaceae), *M. nazimiyensis* from Tunceli, Turkey. Ot Sist. Bot. Dergisi **26**: 13–16.
- Yıldırım, S. and Selvi, B. 2002. A new species, *Muscari sivrihisardaghlarensis* (Liliaceae) from Central Anatolia. Ot Sist. Bot. Dergisi **9**: 7–12.
-