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## Chromosome counts in some Anatolian species of *Mentha* (*Labiatae*)

### Abstract

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Chromosome counts of the species of *Mentha* L. (*Labiatae*) (mint) found in the North Anatolian region of Turkey (A3-A9) are reported. The following chromosome numbers are new counts to Turkish *Mentha*: *Mentha pulegium* L.  $2n=20$ , *M. aquatica* L.  $2n=96$ , *M. suaveolens* Ehrh.  $2n=24$ , *M. longifolia* (L.) Hudson subsp. *longifolia*  $2n=24$ ,  $2n=48$ , *M. longifolia* (L.) Hudson subsp. *typhoides* (Briq.) Harley var. *typhoides*  $2n=24$ , *M. spicata* L. subsp. *spicata*  $2n=48$ , *M. ×dumetorum* Schultes  $2n=60$ , *M. ×villosa-nervata* Opiz  $2n=36$ .

### Introduction

The *Labiatae* (*Lamiaceae*) consists of 200 genera and 3500 species in the world (Kokkini 1994). *Mentha* (mint) is a member of the subfamily *Nepetoideae* and the tribe *Mentheae*. It is grouped into five sections (Harley & Brington 1977; Kokkini 1991).

The family is represented in Turkey by 45 genera and more than 546 species (Seçmen & al. 1992). Species of *Mentha* L. in Turkey represent of these two sections, four hybrids and fourteen taxa. There are eight taxa in North Anatolia two of which are hybrids. These taxa are *M. pulegium*, *M. aquatica*, *M. suaveolens*, *M. longifolia* subsp. *longifolia*, *M. longifolia* subsp. *typhoides* var. *typhoides*, *M. spicata* subsp. *spicata*, *M. ×dumetorum*, *M. ×villosa-nervata*.

Some karyological studies of the *Mentha* species have been reported in the literature. Harley & Brington (1977) studied the chromosome numbers of *M. microphylla*, *M. pulegium*, *M. suaveolens*, *M. longifolia* in England. Tucker and Fairbrothers (1981) determined the chromosome numbers in  $F_1$  progeny of *M. ×gracilis* (*M. arvensis* × *M. spicata*) and later on (1990) studied the chromosome number, fertility and morphological characteristics of this hybrid. A somatic chromosome number of  $2n=24$  for *M. longifolia* has been reported in Italy by Maffei (1988).

Tyagi and Ahmad (1989) studied interspecific somatic chromosome variation in an  $F_1$  hybrid progeny of a cross between *M. spicata* and *M. ×piperita*. Ahmad & al. (1992) studied the variation of nuclear DNA content in *M. ×piperita*, *M. viridis*, *M. spicata* and *M. cardiaca*. Chambers & al. (1994) counted chromosome numbers of *M. australis*, *M. japonica*, *M. diemanica* and *M. cunninghamii* at the USDA-ARS National Clonal

Germplasm Repository. Chromosome numbers of the genus *Mentha* in Turkey have not yet been studied.

Most of the *Mentha* species are characterized by large morphological variations as reflected by high number of names at different taxonomic ranks attributed by taxonomists to mints during the past 200 years. Furthermore, intra and interspecific hybridization occurs commonly when the species of the section *Mentha* meet sympatric condition with each other, leading to the complex variation pattern that characterizes most wild populations.

Apart from their high morphological variability, most mint species are characterized by a great chemical diversity with respect to their essential oil constituents (Kokkini 1994). This study has been carried out to determine the taxonomic problems of eight taxa of *Mentha* spread in North Anatolia.

## Material and methods

*Mentha pulegium*, *M. aquatica*, *M. ×dumetorum*, *M. suaveolens*, *M. longifolia* subsp. *longifolia*, *M. longifolia* subsp. *typhoides* var. *typhoides*, *M. spicata* subsp. *spicata*, *M. ×dumetorum*, *M. ×villosa-nervata* were used in this study. They were collected from Black Sea Region of Turkey during August-October of 1993-1996. Voucher specimens are deposited in the herbarium at the Uludağ University, Faculty of Science, Department of Biology (BULU).

Somatic chromosome counts were determined by a method adapted from Sharma & Bhattachayya (1959), Sharma & Sharma (1972). Root tips 1 cm long were incubated in saturated p-dichlorobenzene in tap water for three hours at 10 °C and then immediately hydrolyzed and stained for six hours in a solution of 9 parts 2% orcein in 45 % acetic acid to 1 part 1 N HCl. at 22 °C. The terminal 1-2 mm section of the root was destained and squashed in 45 % acetic acid (Tucker & Fairbrothers 1990).

The hand drawings shown in Figures 1-9 were taken from Zeiss Standard axioplan light microscope at 1000 × magnification. The chromosome numbers of the eight *Mentha* taxa investigated are shown in Table 1.

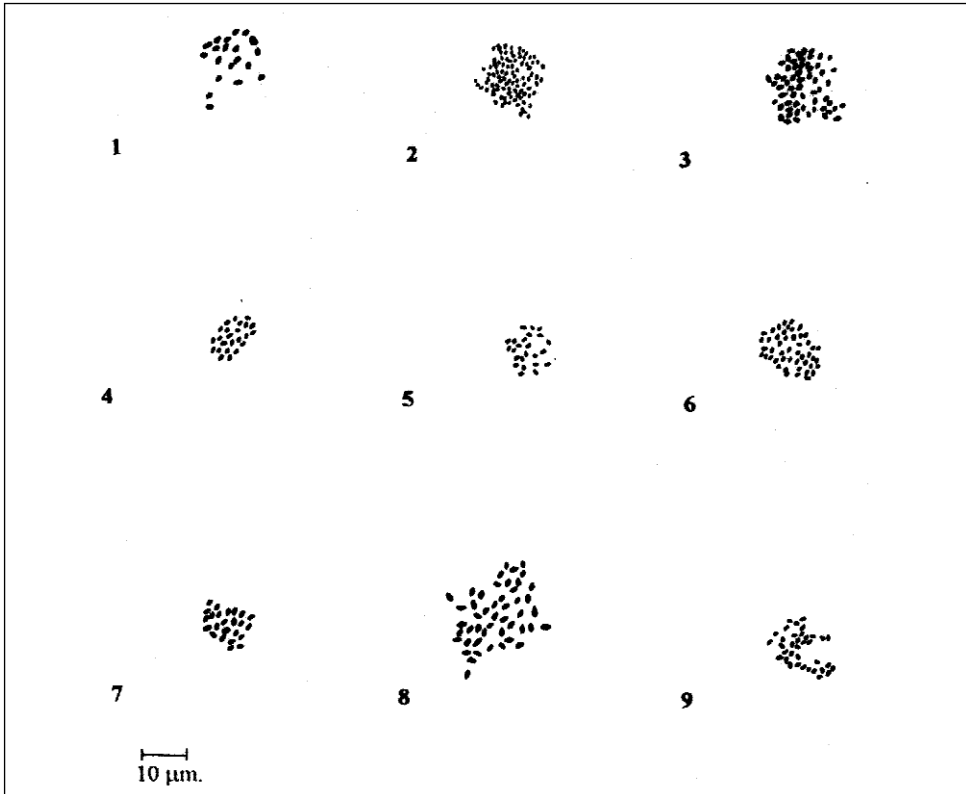
## Results

In the present study, the chromosome numbers of eight *Mentha* taxa, two of which were new records for Turkey, were reported. *M. ×dumetorum* and *M. ×villosa-nervata* are new records.

### Sect. *Pulegium*

#### *Mentha pulegium* L.

It grows in river and lake sides, grass fields, damp places and road sides from sea level up to 2200 m. All collections have a somatic chromosome number of  $2n=20$  (Table 1, Fig. 1).



Figs.1-9. Somatic chromosome numbers at metaphase of *Mentha* species. 1. *M. pulegium*  $2n=20$ ; 2. *M. aquatica*  $2n=96$ ; 3. *M. \times dumetorum*  $2n=60$ ; 4. *M. suaveolens*  $2n=24$ ; 5-6. *M. longifolia* subsp. *longifolia*  $2n=24$ ,  $2n=48$ ; 7. *M. longifolia* subsp. *typhoides* var. *typhoides*  $2n=24$ ; 8. *M. spicata* subsp. *spicata*  $2n=48$ ; 9. *M. \times villosa-nervata*  $2n=36$ .

### Sect. *Mentha*

#### *Mentha aquatica* L.

It grows in river, lake and stream sides, streambeds, rice field, marshes, damp regions that dry up in summer, from sea level up to 1350 m. All collections have a somatic chromosome number of  $2n=96$  (Table 1, Fig. 2).

#### *Mentha \times dumetorum* Schultes

It grows in stream and lake sides, road sides, from 20 to 1450 m. All collections have a somatic chromosome number of  $2n=60$  (Table 1, Fig. 3).

#### *Mentha suaveolens* Ehr.

*M. suaveolens* is a Mediterranean element. The species is seen in damp places, from 5 to 150 m. The single collection examined has a somatic chromosome number of  $2n=24$  (Table 1, Fig. 4).

Table 1. Localities and Chromosome numbers in *Mentha* species collected in North Anatolian Region of Turkey.

Taxon	2n	Localities
<i>Mentha pulegium</i> L.	20	Bartın; Kozcağız, road sides, 100 m., 27.08.1994, G. Tarımcılar, G. Kaynak, BULU 9148.
.	20	Sinop; 2 km. above Ayancık, grassfield, 110m., 26.08.1994, G. Kaynak, G. Tarımcılar, BULU 9013.
	20	Samsun; Kahyalı, E. Of Köprübaşı, road sides, 0 m., 24.08.1993, G. Tarımcılar, G. Kaynak, BULU7718.
	20	Ordu; Ünye, Göbünalcı village, damp places, 10 m.,24.08.1993, G. Kaynak, G. Tarımcılar,BULU 7722.
	20	Ordu; Gürgentepe, road sides, 1020 m., 24.08.1993 G. Tarımcılar, G. Kaynak, BULU 7749.
	20	Giresun; Eşpiye, Armelet village, road sides, 370 m., 26.08.1993, G. Kaynak, G. Tarımcılar, BULU 7793.
	20	Giresun; Görele, road sides, 40 m., 26.08.1993, G. Tarımcılar, G. Kaynak, BULU 7801.
	20	Rize; Çamlıhemşin, road sides, 140 m., 28.08.1993, G. Kaynak, G. Tarımcılar, BULU 7863.
	20	Artvin; Artvin to Borçka, 26 km. below Borçka, 230 m., 29.08.1993, G. Tarımcılar, G. Kaynak, BULU 7901.
<i>Mentha aquatica</i> L.	96	Kastamonu; Hanönü, rice field, 375m., 24.08.1994, G. Kaynak, G. Tarımcılar, BULU 8965.
	96	Kastamonu; Şenpazar to Cide, around Harmangeriş village, damp places, 375 m., 27.08.1994, G. Tarımcılar, G. Kaynak, BULU 9097.
	96	Çorum; Kargı to Osmancık, road sides, 500 m., 24.08.1994, G. Kaynak, G. Tarımcılar,BULU 8982.
	96	Samsun; Taşova to Ladik, lake side, 860 m., 24.08.1994, G. Tarımcılar, G. Kaynak, BULU 9001.
	96	Ordu; Göbünalcı village, damp places, 10 m., 24.08.1993, G. Kaynak, G. Tarımcılar, BULU 7721.
	96	Giresun; Giresun to Dereci, Taşocağı , damp places, 90 m., 25.08.1993, G. Tarımcılar, G. Kaynak, BULU 7751.
	96	Giresun; Eşpiye, Armelet village, road sides, 370 m., 25.08.1993, G. Kaynak, G. Tarımcılar, BULU 7792.
	96	Trabzon; Trabzon to Bayburt, Maçka, road sides, 400 m., 26.08.1993, G. Tarımcılar, G. Kaynak, BULU 7802.
	96	Rize; Güneyce, damp places, 0 m., 27.08.1993, G. Kaynak, G. Tarımcılar, BULU 7842.
	96	Artvin; Ardanuç to Kutul, 24, damp places, 1350 m., 29.08.1993, G. Tarımcılar, G. Kaynak, BULU 7890.
<i>Mentha suaveolens</i> Ehr.	24	Sakarya; Sakarya to Karasu, Limandere, damp places, 50-70 m., 29.09.1995, G. Kaynak, G. Tarımcılar, BULU 9425.
<i>M. longifolia</i> (L.) Hudson subsp. <i>longifolia</i>	24	Bolu; Abant lake, 1350 m., 01.10.1995, G. Tarımcılar, BULU 9481.
	24	Zonguldak; Karabük, Eskipazar, Çay village, stream sides, 1450 m., 21.08.1994, G. Kaynak, G. Tarımcılar, BULU 8896.

Table 1. Continued.

	48	Zonguldak; Yedigöller, 500 m., 27.08.1994, G. Tarımcılar, G. Kaynak, BULU 9180.
	24	Kastamonu; Daday to Azdavay, road sides, 1400 m., 22.08.1994, G. Kaynak, G. Tarımcılar, BULU 8916.
	24	Kastamonu; Daday to Azdavay, Sovucaova, road sides, 1380 m., 22.08.1994, G. Tarımcılar, G. Kaynak, BULU 8924.
	24	Kastamonu; Tosya to Çiftlik, Şahinçatı, damp places, 1200 m., 23.08.1994, G. Kaynak, G. Tarımcılar, BULU 8953.
	24	Kastamonu; Hanönü, streamsides, rice field, 375 m., 24.08.1994, G. Tarımcılar, G. Kaynak, BULU 8966.
	24	Kastamonu; Ağılı to Cide, Hıdırlar village, stream sides, 790 m., 27.08.1994, G. Kaynak, G. Tarımcılar, BULU 9080.
	24	Kastamonu; Cide to Kurucuşile, damp places, 150 m., 27.08.1994, G. Tarımcılar, G. Kaynak, BULU 9135.
	24	Samsun; Tekkeköy cave, road sides, 0 m., 24.08.1994, G. Kaynak, G. Tarımcılar, BULU 7717.
	24	Ordu; Çamaş, Güngören, road sides, 700 m., 24.08.1994, G. Tarımcılar, G. Kaynak, BULU 7736.
	24	Ordu; Kırıktepe, road sides, 900 m., 24.08.1994, G. Kaynak, G. Tarımcılar, BULU 7745.
	24	Ordu; Gürgentepe, road sides, 1030 m., 24.08.1994, G. Tarımcılar, G. Kaynak, BULU 7749.
	24	Rize; İkizdere to Kalkandere, around Çamlık village, road sides, 1550 m., 27.08.1993, G. Kaynak, G. Tarımcılar, BULU 7836.
	24	Artvin; Ardanuç to Kutul, Harmanlı village, road sides, 720 m., 29.08.1993, G. Tarımcılar, G. Kaynak, BULU 7889.
	24	Artvin; Artvin to Kutul, 1000 m., 29.08.1993, G. Kaynak, G. Tarımcılar, BULU 7889A.
<i>M. longifolia</i> (L.) Hudson subsp. <i>typhoides</i> (Briq.) Harley var. <i>typhoides</i>	24	Zonguldak; Zonguldak to Muslu, 36 m., 30.09.1995, G. Tarımcılar, G. Kaynak, BULU 9449.
	24	Bartın, road sides, 25 m., 27.08.1994, G. Kaynak, G. Tarımcılar, BULU 9146.
	24	Kastamonu; Ağılı to Cide, Şenpazar, road sides, 500 m., 27.08.1994, G. Tarımcılar, G. Kaynak, BULU 9095.
	24	Sinop; Ayancık, Yenikonak, road sides, 215m., 26.08. 1994, G. Kaynak, G. Tarımcılar, BULU 9022.
	24	Samsun; Çarşamba, Kutlukent, road sides, 0m., 24.08.1993, G. Tarımcılar, G. Kaynak, BULU 7716.
	24	Amasya, road sides, 400 m., 25.08.1994, G. Kaynak, G. Tarımcılar, BULU 8985.
	24	Ordu; Ünye, Göbünalcı village, near field, 10m., 24.08.1993, G. Tarımcılar, G. Kaynak, BULU 7720.
	24	Ordu; Fatsa to Aybastı, Gaga lake sides, 85 m., 24.08.1993, G. Kaynak, G. Tarımcılar, BULU 7724.
	24	Trabzon; Trabzon to Maçka 16 km., streambed that dry up in summer, 440 m., 26.08.1993, G. Tarımcılar, G. Kaynak, BULU 7802.
	24	Rize; Çaykara, damp place, 430 m., 27.08.1993, G. Kaynak, G. Tarımcılar, BULU 7830 A.

Table 1. Continued.

	24	Artvin; Artvin to Düzlüce, stream sides, 240m., 28.08.1993, G. Tarımcılar, G. Kaynak, BULU 7883.
<i>M. spicata</i> L. subsp. <i>spicata</i>	48	Kastamonu; Araç to Daday, edge of forest ( <i>Pinus</i> sp. <i>Ulmus</i> sp., <i>Quercus</i> sp.), 915 m., 22.08.1994, G. Kaynak, G. Tarımcılar, BULU 8905.
	48	Kastamonu; Daday to Azdavay, edge of <i>Abies</i> sp. forest, 1350 m., 22.08.1994, G. Tarımcılar, G. Kaynak, BULU 8914.
	48	Kastamonu; Daday to Azdavay, around Sovucaova, road sides, 1370 m., 22.08.1994, G. Kaynak, G. Tarımcılar, BULU 8922.
<i>Mentha x dumetorum</i> Schultes	60	Sakarya; Sakarya to Karasu, Limandere, damp places, 70 m., 29.09.1995, G. Tarımcılar, BULU 9428.
	60	Bolu; Abant lake side, 1350 m., 01.10.1995, G. Tarımcılar, BULU 9431.
	60	Zonguldak; Karabük to Eskipazar, Çay village, stream side, 1450 m., 21.08.1994, G. Kaynak, G. Tarımcılar, BULU 8837.
<i>Mentha x villosa-nevata</i> Opiz	36	Kastamonu; Daday to Azdavay, edge of <i>Abies</i> forest, 1300 m., 22.08.1994, G. Tarımcılar, G. Kaynak, BULU 8913.
	36	Kastamonu; Daday to Azdavay, around Sovucaova, road sides, 1370 m., 22.08.1994, G. Kaynak, G. Tarımcılar, BULU 8926.

### *Mentha longifolia* (L.) Hudson subsp. *longifolia*

This species is a Euxine element. It grows in lake and stream sides, rice fields, streambeds, marshy and grass fields and damp regions that dry up in summer, from sea level up to 2300 m. The somatic chromosome number of fifteen samples was  $2n=24$ , but the somatic chromosome number of one sample was  $2n=48$ . This plant was collected from Zonguldak; Yedigöller at 500 m. (Table 1, Figs 5-6).

### *Mentha longifolia* (L.) Hudson subsp. *typhoides* (Briq.) Harley var. *typhoides*

It grows in stream and lake sides, rice fields, marshy and grass fields, streambeds that dry up in summer, under *Coryllus* sp., *Rubus* sp., sides of *Fagus* sp. and *Abies* sp. forest from the sea level up to 2140 m. The somatic chromosome number of all samples was  $2n=24$  (Table 1, Fig. 7).

### *Mentha spicata* L. subsp. *spicata*

It grows in stream sides, damp places, ditch and road sides from the sea level up to space 220 m. The somatic chromosome number of all samples was  $2n=48$  (Table 1, Fig. 8).

### *Mentha x villosa-nervata* Opiz

It grows in stream and road sides and dry soil, from the sea level up to 1400 m. The chromosome number of both samples was  $2n=36$  (Table 1, Fig. 9).

## Discussion

The counts of somatic chromosome numbers agree with the previous reports;  $2n=20$  of *M. pulegium* (diploid) (Davis 1982).  $2n=20$  and  $2n=40$  (tetraploid) ( Harley & Britton

Table 2. Chromosome counts in *Mentha*: Somatic numbers, base numbers, origin of material.

Taxon	Chromosome number	Base Number	Origin of Material	References
<i>Mentha pulegium</i>	20	10	Turkey	Davis 1982
	20	10	England, Greece, Bulgaria,	Harley, Brighton 1977,
	40	10	Morocco	
	20, 40	10	Portugal	
			Canada, U.S.A.	Lawrence 1981
	30	10	England, Tunisia,	Chambers and Hummer 1994
			Australia, Chile, U.S.A.,	
			Spain	
	20	10	Turkey	Tarımcılar and Kaynak 1998
<i>M. aquatica</i>	96	12	Turkey	Davis 1982,
	96	12	Netherlands	Chambers and Hummer 1994
	60, 84, 108	12	Canada, U.S.A.	Lawrence 1981
			Europa	Harley 1977
	96	12	Turkey	Tarımcılar and Kaynak 1998
<i>M. x dumetorum</i>	60	12	Canada	Lawrence 1981
	60	12	Turkey	Tarımcılar and Kaynak 1981
<i>M. suaveolens</i>	24	12	Turkey	Davis 1982
	36, 48	12	Canada, U.S.A.	Lawrence 1981
	24	12	France, Netherlands,	Chambers and Hummer 1994
			Bulgaria, Portugal, U.S.A.	
	24	12	Turkey	Tarımcılar and Kaynak 1998
<i>M. longifolia</i> subsp. <i>longifolia</i>	24, 48	12	Italy	Maffei 1988
	24	12	Netherland, France,	Chambers and Hummer 1994
			Uzbekistan	
	48	12	Afghanistan	Chambers and Hummer 1994
	18, 24, 36, 48	12	Europa, Asia, Africa,	Harley and Brington 1977,
			Canary Islands, Northern	
			and Western Europa	
			Canada	Lawrence 1981

Table 2. Continued.

	18, 24, 36, 48	12	Europa, Asia, Africa, Canary Islands, Northern and Western Europa Canada	Harley and Brington 1977,    Lawrence 1981
	24, 48	12	Turkey	Tarımcılar and Kaynak 1998
<i>M. longifolia</i> subsp. <i>typhoides</i> var. <i>typhoides</i>	24	12	Turkey	Tarımcılar and Kaynak 1998
<i>M. spicata</i> subsp. <i>spicata</i>	48	12	Europa	Harley and Brington 1977
	24, 36, 48, 54	12	Europa, North America	Lawrence 1981
	48	12	Greece	Kokkini and Vokou 1989
	48	12	Czechoslovakia, U.S.A., U.K.	Tucker and Fairbrothers 1990
	36, 48	12	India	Tausif and Tyagi 1992
	36	12	U.S.A	Chambers and Hummer 1994
	48	12	Australia, Italy, Netherlands	Chambers and Hummer 1994
	48	12	Turkey	Tarımcılar and Kaynak 1998
<i>M. xvillosa-nervata</i>	36	12	Greece	Kokkini and Papageorgiou 1987, Kokkini and Vokou 1989
	36	12	Turkey	Tarımcılar and Kaynak 1998



1977; Lawrence 1981). In these studies, it is reported that diploid and tetraploid *M. pulegium* have similar morphological structures although they have different chromosome numbers. A chromosome number of  $2n=20$  (diploid) for *M. pulegium* from England, Tunisia, Australia, Chile, U.S.A (Los Angeles, Oregon) and Spain (Cordoba) contrasts with reports of  $2n=30$  (triploid) based on plants from the U.S.A (Oregon) and Poland (Chambers & Hummer 1994). Our counts of *M. pulegium* are all at the diploid level based on (Table 1, Fig. 1).

In this study the count for *M. aquatica* was found to be  $2n=96$  (Table 1, Fig. 2). Earlier counts for *M. aquatica* are  $2n=96$  (octoploid; Davis 1982; Chambers & Hummer 1994),  $2n=60$  (pentaploid),  $2n=84$  (heptaploid),  $2n=108$  (nonaploid) (Lawrence 1981; Harley 1977). Based on known counts for the species, a base number of  $x=12$  appears probable.

In the study of Lawrence (1981) the count for *M. × dumetorum* was reported to be  $2n=72$  (heptaploid) and  $2n=84$  (hexaploid). However it was stated that a count  $2n=60$  (pentaploid) should be present. In this study, their statement is validated, it was found for *M. × dumetorum* that  $2n=60$  with a base number  $x=12$  (Table 1, Fig. 3).

For our sample of *M. suaveolens*, the chromosome number was  $2n=24$  (diploid) with a base number of  $x=12$  (Table 1, Fig. 4). Earlier counts in the literature were  $2n=24$  by Davis (1982) and  $2n=24$  (diploid),  $2n=36$  (triploid),  $2n=48$  (tetraploid) by Lawrence (1981). Whereas for the samples collected by Chambers & Hummer (1994) in France, Netherlands, Bulgaria, Portugal, U.S.A (Minnesota, Minneapolis, Oregon, Marion) the count was  $2n=24$  (diploid).

Our chromosome count for one sample of *M. longifolia* subsp. *longifolia* from Zonguldak was  $2n=48$  (tetraploid) and other collections were  $2n=24$  (diploid) with a base number of  $x=12$  (Table 1, Figs 5-6). Previously published counts by Chambers & Hummer (1994) for *M. longifolia* subsp. *longifolia* from The Netherlands, France (Dijon) and Uzbekistan (Tashkent) were  $2n=24$  (diploid), whereas those from Afghanistan (Kataghan, Herat) are  $2n=48$  (tetraploid). These counts were consistent with the findings of Harley & Brington (1977) and Lawrence (1981), who reported  $2n=18$ ,  $2n=24$ ,  $2n=36$ ,  $2n=48$  and those of Maffei (1988) who reported  $2n=24$ ,  $2n=48$ . In our studies *M. longifolia* subsp. *typhoides* var. *typhoides* was diploid ( $2n=24$ ) with a base number of  $x=12$  (Table 1, Fig. 7).

Our chromosome counts of *M. spicata* subsp. *spicata* were  $2n=48$  (tetraploid) with a base number of  $x=12$ . In other studies, chromosome counts of *M. spicata* subsp. *spicata* have been reported as  $2n=48$  (tetraploid) by Harley & Brington (1977);  $2n=24$  (diploid),  $2n=36$  (triploid),  $2n=48$  (tetraploid),  $2n=54$  (hypo-pentaploid) by Lawrence (1981);  $2n=48$  (tetraploid) by Kokkini & Vokou (1989);  $2n=48$ , by Tucker & Fairbrothers (1990) and  $2n=36$  and  $2n=48$  by Ahmad & al. (1992). Chambers & Hummer (1994) have reported  $2n=36$  for samples of *M. spicata* collected in U.S.A (Florida) and  $2n=48$  for glabrous-leaf samples collected from Australia, Italy, Netherlands, U.S.A (Pennsylvania). They have studied samples with rugose-leaf forms collected in U.S.A (Washington, New Mexico, California, Florida), Australia, England, Brazil are found  $2n=48$ . Their samples included hairy-leaf and crisp-leaf forms all with  $2n=48$ .

For the other hybrid species of the research area, *M. × villosa-nervata*, the chromosome count showed  $2n=36$  (triploid) with a base number of  $x=12$  (Table 1). Kokkini and Papageorgiou (1987) and Kokkini and Vokou (1989) also reported previously  $2n=36$  in the same species collected in Greece.

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