



## New chromosome numbers in the genus *Marrubium* (Horehound) from Turkey

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### Abstract

In this research, the chromosome numbers of six taxa of the genus *Marrubium* L. (Horehound) growing naturally in Turkey were counted. Of the taxa of the genus *Marrubium* (Lamiaceae), *Marrubium vulgare* L. has somatic chromosome number of  $2n=34+2B$ , *M. vulcanicum* Hub.-Mor.  $2n=32$ , *M. bourgaei* Boiss. subsp. *bourgaei*  $2n=30$ , *M. bourgaei* Boiss. subsp. *caricum* P.H. Davis and *M. astracanicum* subsp. *astracanicum* Jaq.  $2n=20$ , *M. peregrinum* L.  $2n=34$ . Chromosome numbers of the three taxa examined are presented for the first time.

**Key words:** Chromosome number, *Marrubium*, Lamiaceae, Turkey

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### Türkiye'den *Marrubium* (Köpek otu) cinsinde yeni kromozom sayıları

#### Özet

Bu araştırmada, Türkiye'de doğal olarak yetişen *Marrubium* L. (Köpek otu) cinsine ait altı taksonun kromozom sayısı sayıldı. *Marrubium* (Lamiaceae) cinsine ait taksonların sahip olduğu somatik kromozom sayıları, *Marrubium vulgare* L.  $2n=34+2B$ , *M. vulcanicum* Hub.-Mor.  $2n=32$ , *M. bourgaei* Boiss. subsp. *bourgaei*  $2n=30$ , *M. bourgaei* Boiss. subsp. *caricum* P.H. Davis ve *M. astracanicum* subsp. *astracanicum* Jaq.  $2n=20$ , *M. peregrinum* L.  $2n=34$ 'dür. İncelenen taksonlardan üç tanesinin kromozom sayısı ilk kez sunulmaktadır.

**Anahtar kelimeler:** Kromozom sayı, *Marrubium*, Lamiaceae, Türkiye

#### 1. Introduction

*Marrubium* L. (Horehound) contains herbaceous plants distributed in the Irano-Turanian and Mediterranean phytogeographic regions (Hedge, 1992). The total number of taxa is about 40. Twelve are recorded in Europe (Cullen, 1972), 14 in the former USSR, (Komarov, 1954) and 15 in Iran (Seybold, 1978). In Turkey there are 21 species, with one subspecies and six varieties (Akgül et al., 2008).

The genus was first revised by Bentham (1834, 1848), who divided it into two sections, *Lagopsis* and *Marrubium*. Later, the taxonomy of the genus was treated by several workers and the genus was divided into various sections on the basis of morphological characters: three sections (*Ballatoides*, *Marrubium* and *Lagopsis*) by Briquet (1896), two sections (*Eumarrubium* and *Ballatoides*) by Boissier (1879) and four sections (*Marrubium*, *Afghanica*, *Stellata* and *Microdontha*) by Seybold (1978). On the other hand, Grossheim (1967) had only the name section, and Cullen (1982) and more recently did not assign the Turkish species to any sections (Akgül et al., 2008).

The somatic chromosome numbers of *Marrubium alternidens*, *M. anisodon*, *M. astracanicum*, *M. incanum*, *M. leonuroides*, *M. peregrinum*, *M. supinum*, *M. thessalum*, *M. velutinum* and *M. vulgare* in the genus *Marrubium* were also reported as  $2n=30$ , 32 and 34 (Löve and Kjellqvist 1974; Van Loon and Kieft, 1980; Astanova, 1981; Strid and

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Franzen, 1981; Gill, 1981, 1983; Queiros, 1983; Fernandes and Leitão, 1984; Magulaev, 1984; Rosúa and Navarro, 1986; Pogan et al., 1989; Luque and Díaz Lifante 1991; Baltisberger, M. 1991a, b, c; Ruíz de Clavijo Jiménez, 1994; Baltisberger M. and E. Baltisberger, 1995; Dobeš et al., 1996, 1997; Murín, 1997; Baltisberger, 1999; Valdés et al., 1999).

In this study we present somatic chromosome numbers of six taxa of this important genus in Lamiaceae. Of the examined taxa, three taxa were identified karyologically for the first time.

## 2. Materials and methods

The surface of seeds was abraded with emery paper to accelerate germination. Seeds were germinated on moist filter papers in Petri dishes at room temperature in a very short time. Root tips were pretreated with  $\alpha$ -monobromonaphthalene at 4 °C for 16 h and fixed with Carnoy for 24 h at 4 °C. Before staining, the material was hydrolyzed with 1N HCl for 12 minutes at room temperature. The chromosomes were stained with 2% acetic orcein and mounted in 45% acetic acid (Martin et al., 2011). Permanent slides were made by using the standard liquid nitrogen method. The somatic chromosome number of each taxon was counted by the use of permanent preparations containing chromosomes in the metaphase stage of mitosis. Photographs were taken at 10 × 100 magnification under a light microscope and transferred to a computer screen after detecting somatic cells with well-spread chromosomes without shrinkage. The somatic chromosome numbers were counted considering the enlarged micrographs of ten well-spread metaphase plates. Also, they were calculated with the Bs200Pro Image Analysis System (Çetin et al., 2010).

## 3. Results

Of the six taxa of the genus *Marrubium* whose somatic chromosome numbers were determined by this study, the taxa of *Marrubium vulcanicum*, *M. bourgaei* subsp. *bourgaei* and *M. bourgaei* subsp. *caricum* are endemic to Turkey.

*Marrubium vulgare* L.

Location: C4 Karaman: Konya-Karaman entrance, 650-750 m., 21.06.2001, roadside, Akgül 2455. Somatic chromosome number:  $2n=34+2B$  (Figure 1a).

*Marrubium vulcanicum* Hub.- Mor.

Location: B9 Ağrı: Patnos, through Patnos to Erçiş 3. km, on the right side of the road, 1700 m., 19.09.2001, rocky slopes, around volcanic rocks, Akgül 2525. Somatic chromosome number:  $2n=32$  (Figure 1b).

*Marrubium bourgaei* Boiss. subsp. *bourgaei*

Location: C2 Burdur: Altınyayla (Dirmil gateway), Kırkpınar Fatmapınar plateaux, 1700-1800 m., 30.09.2001. Somatic chromosome number:  $2n=30$  (Figure 1c).

*Marrubium bourgaei* Boiss. subsp. *caricum* P.H. Davis

Location: C2 Denizli: Acıpayam, above Ören village, Bozdağ Mount, near the peak, 2000-2100 m. 29.09.2001, limestone. Somatic chromosome number:  $2n=20$  (Figure 1d).

*Marrubium peregrinum* L.

Location: B3 Eskişehir: Türkmen Mount, Aşağı Kalabak, 950 m., 26.07.1997, Akgül 2646. Somatic chromosome number:  $2n=34$  (Figure 1e).

*Marrubium astracanicum* subsp. *astracanicum* Jaq.

Location: C5 Niğde: Çamardı, the Aladağlar Mount, above the camping area, 1800 m, 05.07.2002, rocky openings, along with the communities of *Juniperus* sp., *Astragalus* sp., *Berberis vulgaris* sp. Somatic chromosome number:  $2n=20$  (Figure 1f).

## 4. Discussion

Chromosome studies were performed on the taxa of *Marrubium alternidens*, *M. anisodon*, *M. astracanicum*, *M. incanum*, *M. leonuroides*, *M. peregrinum*, *M. supinum*, *M. thessalum*, *M. velutinum* and *M. vulgare* (Löve and Kjellqvist 1974; Van Loon and Kieft, 1980; Astanova, 1981; Strid and Franzen, 1981; Gill, 1981, 1983; Queiros, 1983; Fernandes and Leitão, 1984; Magulaev, 1984; Rosúa and Navarro, 1986; Pogan et al., 1989; Luque and Díaz Lifante, 1991; Baltisberger, M. 1991a, b, c; Ruíz de Clavijo Jiménez, 1994; Baltisberger M. and E. Baltisberger, 1995; Dobeš et al., 1996, 1997; Murín, 1997; Baltisberger, 1999; Valdés et al., 1999). Our results obtained for the species of *Marrubium astracanicum*, *M. peregrinum* and *M. vulgare* are inconsistent with the previous studies resulting from the difference in locality. According to the literature, *Marrubium vulgare* has a somatic chromosome number of  $2n=34$  (Rosúa and Navarro, 1986; Luque and Díaz Lifante, 1991). In our study, following the repeated preparations, 2B chromosomes were observed for this species. These chromosomes are presented in the photograph (Figure 1a).

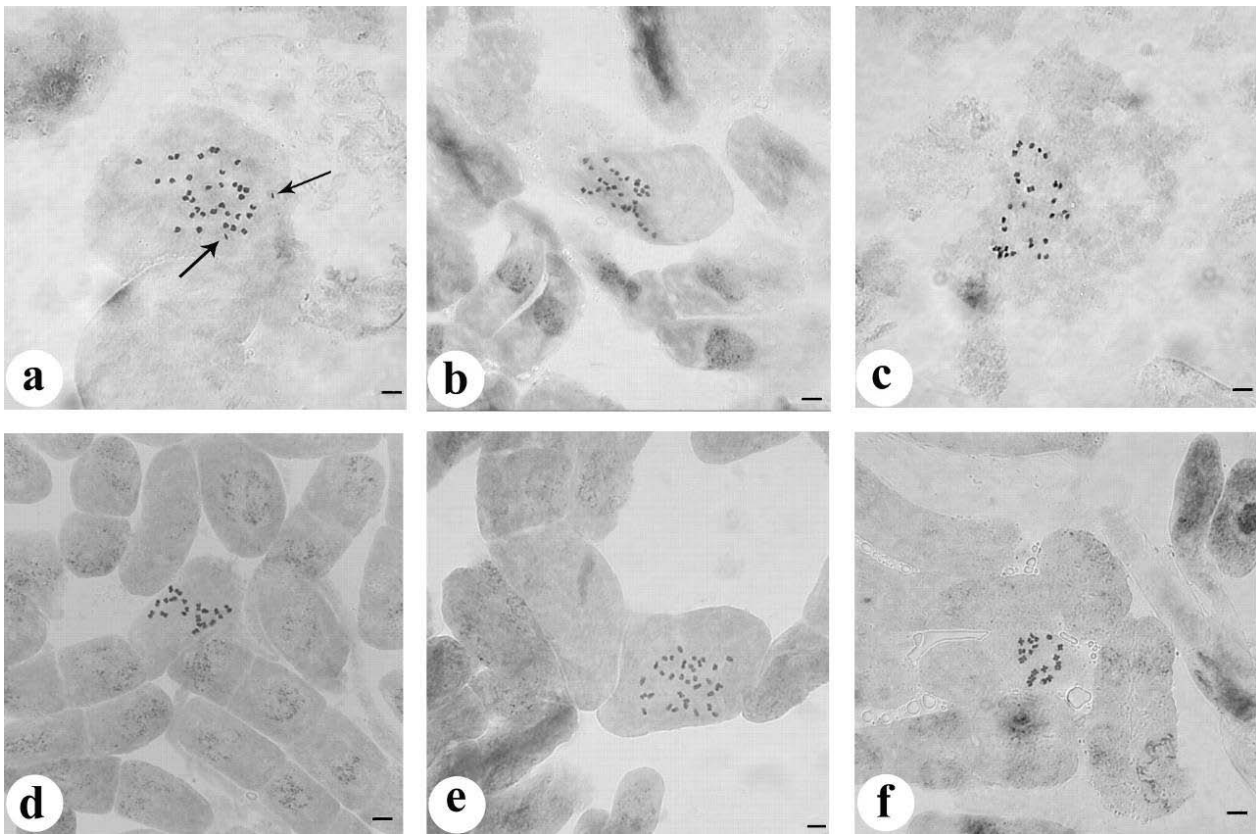


Figure 1. Somatic chromosomes of *Marrubium* taxa a) *M. vulgare*  $2n=34+2B$ , b) *M. vulcanicum*  $2n=32$ , c) *M. bourgaei* subsp. *bourgaei*  $2n=30$ , d) *M. bourgaei* subsp. *caricum*  $2n=20$ , e) *M. peregrinum*  $2n=34$ , f) *M. astracanicum* subsp. *astracanicum*  $2n=20$ , Bar = 5  $\mu\text{m}$ .

Various somatic chromosome numbers observed in the species *Marrubium supinum* from previous studies support our results. Löve and Kjellqvist (1974) reported that the species has a somatic chromosome number of  $2n=34$ . Ruíz de Clavijo Jiménez (1994) counted the somatic chromosome number of the species as  $2n=32$  although Luque and Díaz Lifante (1991) stated that the species has  $2n=30$  chromosomes.

For the species of *Marrubium peregrinum*, two different somatic chromosome numbers were reported, i.e.  $2n=32$  and  $2n=34$  (Baltisberger M. and E. Baltisberger, 1995). In our study, we determined that the species have  $2n=34$  somatic chromosomes, consistent with the literature.

Being one of the species whose chromosome number was reported in previous studies (Baltisberger, M. 1991c), *Marrubium astracanicum* is found to have  $2n=30$  chromosomes as in the literature.

In this study, some taxa of the genus *Marrubium* in Lamiaceae such an important and a large family growing naturally in Turkey were examined to obtain somatic chromosome numbers by using squash method for preparation. Karyological results obtained from the study indicate that the genus does not have a stable chromosome number showing various patterns among the taxa. We consider our results will pave the way for further studies on this genus.

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(Received for publication 28 April, 2011; The date of publication 15 August 2011)