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Research Article

NEW CHROMOSOME COUNTS IN LAMIACEAE FROM FLORA OF IRAN - II

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ABSTRACT. The mitotic chromosome counts were presented for five species (including six populations) belonging to four genera of Lamiaceae from south and southeast of Iran. Among them chromosome numbers of *Rydingia persica* and *Lagochilus macracanthus* are reported for the first time. The chromosome counts for *Melissa officinalis* is in accordance with the previous reports. While the chromosome number for *Rydingia michauxii* and *Drepanocaryum sewerzowii* does not confirm the previous counts.

Keywords: *Drepanocaryum, Lagochilus, Melissa, Otostegia, Rydingia.*

INTRODUCTION

Lamiaceae is one of the most important families of the flowering plants, which consists of ca. 240 genera across more than 7200 species with a world-wide distribution [5]. Plants from Lamiaceae are aromatic commonly used for flavoring, fragrance and medical purpose. The flora of Iran comprises approximately 46 genera and 406 species of Lamiaceae including the species of *Drepanocaryum* Pojark., *Lagochilus* Bunge ex Benth., *Melissa* L. and *Rydingia* Scheen & V.A. Albert [7, 13].

Drepanocaryum contains only one known species, *D. sewerzowii* (Regel) Pojark, (syn. *Nepeta sewerzowii* Regel) distributed from Iran to Central Asia and Pakistan [21]. This is an annual and grows primarily in the temperate biome. The plants grow in shady places, under rocks or bushes. This species is easily distinguished by its basally oblique-inflated calyces and the nutlet shape [6]. The nutlets are globose, tuberculate, with large and curved areole [5].

Lagochilus is a genus of tribe Leonureae, subfamily Lamioideae, Lamiaceae [3, 14, 19], distributed in dry slopes, valleys, and deserts of Iran to Mongolia, Russia (S. Siberia), NW China and N. Pakistan [5]. *Lagochilus* is the largest genus within Leonureae, and comprises ca. 40 species. The species have spiny bracteoles longer than calyces, and a densely villous, 2-lobed, long and narrow posterior corolla lip. Spines usually present in the leaf axils [5]. According to Jamzad [7], *Lagochilus* in Iran includes six species distributed mainly in arid areas of the Irano-Turanian region.

Melissa is a genus of perennial herbs in Lamiaceae, tribe Mentheae, and includes four species usually in forests, Europe, N Africa, Macaronesia and Asia. Plants are widely utilized as a source of essential oils, both medicinally and as flavouring [5]. This genus has one species in Iran, namely *M. officinalis* L. [7, 13].

Rydingia belongs to tribe Leucadeae, subfamily Lamioideae, Lamiaceae [3, 5, 19]. Based on morphological and molecular analyses of Scheen and Albert [17, 18], *Otostegia integrifolia* Benth., *O. limbata* (Benth.) Boiss., *O. michauxii* Briq., and *O. persica* (Burm.f.) Boiss. separated from the remaining species of *Otostegia* Benth. Therefore, these four species were transferred to the new genus *Rydingia*. Three species are endemic to Pakistan, Iran and or Western Himalaya, and one species (*Rydingia integrifolia* (Benth.) Scheen & V.A. Albert) is distributed in Eritrea, Ethiopia, and Yemen [6, 13, 20]. All four species have spines at the leaf-axils as well as spinose and persistent bracteoles, yellow flowers and few-flowered verticillasters [6, 13, 15].

In this study, the chromosome numbers of five species belonging to four genera including *Drepanocaryum, Lagochilus, Rydingia* and *Melissa* were examined. This study will help a better understanding of the relationships among the taxa and provide guidance for further investigations in future.

MATERIALS AND METHODS

Plant materials

Materials for chromosome count studies include the species of *Drepanocaryum*, *Lagochilus, Melissa* and *Rydingia*. Selection of the studied species was done according to the materials deposited in MIR herbarium (Table 1). Seeds and herbarium specimens of the studied materials were collected from their habitats in Iran (Fig. 1). Online databases, as well as related literatures, were used to assess the previous chromosomal works on the examined taxa.

Chromosome count investigations

At least ten root tips were collected from numerous germinating seeds of the studied species. The roots were pretreated with α-monobromonaphthalene, 5 h at 4 °C and then fixed overnight in ethanol: glacial acetic acid (3:1). The meristematic tissue was then thoroughly rinsed in distilled water, hydrolysed in 1 N HCl at 60°C for 1-2 min, and stained in aceto-iron hematoxilin for 2 h at 30°C [12]. The meristems were finally dissected and squashed on glass slides in a drop of 45 % acetic acid. Metaphase plates were examined with an OPTIKA – OPTIKAM HDMI – 4083.13 microscope photomicrograph system.

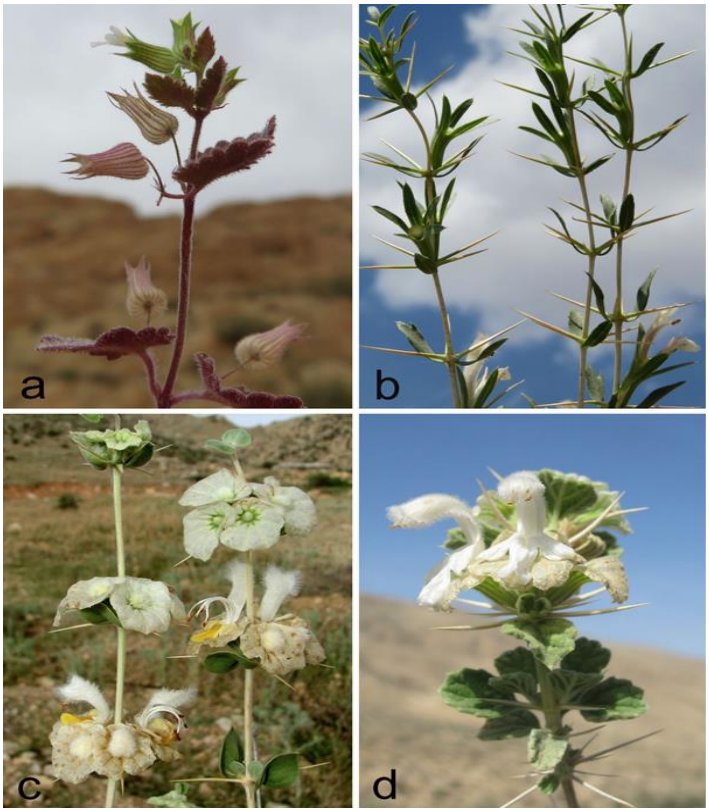

Fig. 1. *Plants of Lamiaceae from flora of Iran. (a) Drepanocaryum sewerzowii (b) Lagochilus macracanthus (c) Rydingia michauxii and (d) Rydingia persica. Photographs: a, b M.Mirtadzadini; c, d F.Bordbar.*

Table 1. *Voucher information and chromosomal data of Lamiaceae from flora of Iran examined in this study.*

Taxon	Voucher information	Chromosome number
Drepanocaryum sewerzowii (Regel) Pojark	Iran: SE, Kerman Prov., NE of Kerman, SE of Fusk village, 30°23'51"N, 57°21'41.7"E, 1975 m, 28.IV.2021, *Mirtadzadini and Bordbar 4160* (MIR)	2n=18
Lagochilus macracanthus Fisch. & C.A.Mey	Iran: SE, Kerman Prov., NE of Kerman, near Gishin, 30°21'35"N, 57°17'50.5"E, 2395 m, 09.VI.2020, *Mirtadzadini and Bordbar 4161* (MIR)	2n=32
Melissa officinalis L.	Iran: SE, Kerman Prov., NNW of Jiroft, W of Magh, 28°58'48.9"N, 57°37'08.1"E, 1815 m, 04 June 2015, *Bordbar 4162* (MIR)	2n=32

Table 1. *(Continues).*

Taxon	Voucher information	Chromosome number
Rydingia michauxii (Briq.) Scheen & V.A.Albert	Iran, S, Fars Prov., between Dasht-e-Arjan and Kazerun, before Kandei village, 29°44.696'N, 51°47.019'E, 1379 m, 21 June 2018, *Bordbar 3848* (MIR)	2n=84
Rydingia persica (Burm.f.) Scheen & V.A.Albert	Iran, SE, Kerman Prov., N of Jiroft, between Bagh-e Alishir and Koldan, 28°54'41.5"N, 57°40'35"E, 06.X.2020, *Bordbar and Mirtadzadini 4163* (MIR)	2n=28
	Iran, S, Fars Prov., Fasa, Tang-e Karam village, 25.IX.2021, *Bordbar 3849* (MIR)	2n=28

RESULTS AND DISCUSSION

In the present study, the chromosome numbers of six populations representing of five species (four genera) of Lamiaceae from south and southeast of Iran (Fig. 1 and 2) are determined, of which chromosome numbers of two species are studied for the first time. One count confirms previous reports, and two represented counts differ from those cited previously. Chromosome numbers and polyploidy level for each genus are discussed below. The somatic chromosome numbers (2n) for the studied populations is presented in Table 1.

Drepanocaryum

Drepanocaryum sewerzowii was reported from northeast and central parts of Iran [7, 13]. Field exploration in Kerman Province revealed the existence of this species in south of Iran as well (Fig. 1a).

The observations of over twenty mitotic metaphase cells strongly indicated 2n=18 for this species (Fig. 2a) while according to Astanova [1], the plants from Tajikistan has 2n=16. Therefore, the result is not in accordance with the previous finding for this species.

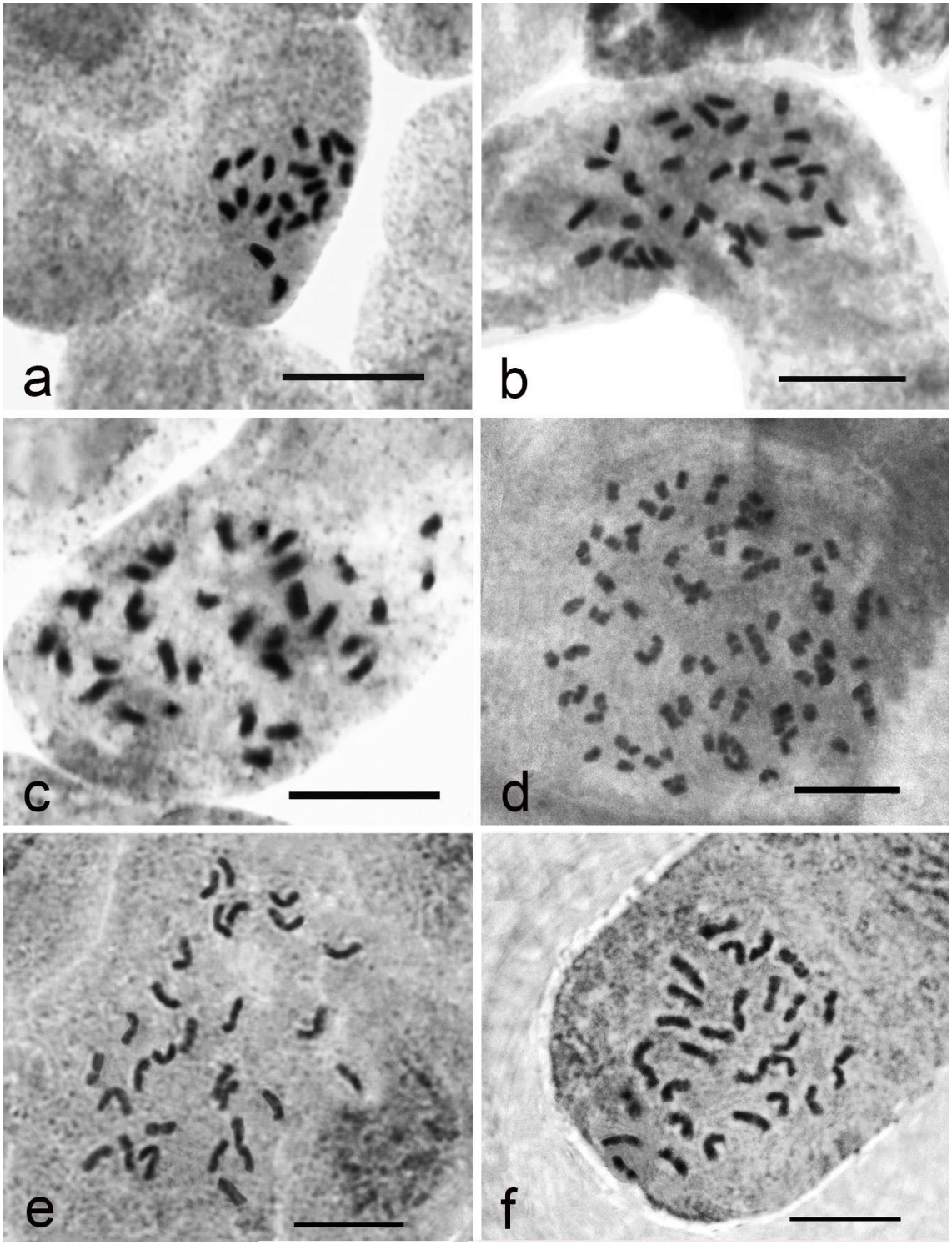

Fig. 2. *Mitotic metaphase chromosomes of Lamiaceae from flora of Iran. (a) Drepanocaryum sewerzowii (Bordbar and Mirtadzadini 4160), (b) Lagochilus macracanthus (Bordbar and Mirtadzadini 4161), (c) Melissa officinalis (Bordbar 4162), (d) Rydingia michauxii (Bordbar 3848), (e) Rydingia persica (Bordbar and Mirtadzadini 4163), (f) Rydingia persica (Bordbar 3849). Scale bar = 10 µm.*

Lagochilus

Lagochilus macracanthus Fisch. & C.A.Mey is an endemic species for the flora of Iran. According to Jamzad [7] this species is distributed in central parts of Iran, and the existence of this species in south of Iran, Kerman Province is confirmed here (Fig. 1b).

The results obtained from the study of several mitotic metaphase cells indicated the chromosome number of 2n= 32 (Fig. 2b). This is the first chromosome number report for this species. According to a summary on Lamiaceae [5], the chromosome number in *Lagochilus* is 2n = 22, and 34. Also, the chromosome number in *L. schugnanicus* Knorring was reported as 2n = 22 [2].

Melissa

Melissa officinalis naturally grows in north, northwest and west of Iran [7, 13], and recently has been reported from south of Iran, Kerman Province as well [4]. This is a medicinal plant cultivated in some parts of Iran. The species has three subspecies; *M. officinalis* ssp. *Officinalis*, ssp. *altissima* (Sm.) Arcang. and ssp. *inodora* Bornm. [13]. Based on the results of Kittler et al. [8], *M. officinalis* ssp. *officinalis* has 2n = 2x = 32 chromosomes and is a diploid whereas ssp. *altissima* with 2n = 4x = 64 chromosomes is a tetraploid. They reported a basic chromosome number of x = 16 for *M. officinalis* for the first time. According to Rechinger [13] the plants from Iran all belong to ssp. *Officinalis*. Based on the observation and in agreement with the previous result, the population from Kerman Province of Iran which also belongs to this subspecies, has 2n=32 chromosome (Fig. 2c).

Rydingia

Three species belonging to *Otostegia* have been reported from the flora of Iran [7, 13]. *O. aucheri* Boiss. in the monotypic *Otostegia* sect. *Mucrophyllon* transferred to *Moluccella* L. and *O. persica* and *O. michauxii* circumscribed as *Rydingia* [17]. *Rydingia persica* (Burm.f.) Scheen & V.A. Albert, a widespread species in the south of Iran is also distributed in SW of Pakistan and N of Oman [6, 11] but *R. michauxii* (Briq.) Scheen & V.A. Albert is a narrow endemic species, only found in Fars Province of Iran (Fig. 1c).
Two populations of *R. persica* from Fars and Kerman Provinces are included in the present study (Fig. 1d). The chromosome count studies revealed 2n = 28 for both populations (Figs. 2e and f). In my knowledge, this is the first chromosome number report for this species. The results also indicated 2n = 84 for *R. michauxii* (Fig. 2d). It seems that *R. persica* and *R. michauxii* are diploid and hexaploid species, respectively, with the basic chromosome number of x= 14. *R. michauxii* was reported to possess 2n = 82 chromosomes by Sadeghian et al. [16] which seems erroneous. The collection area for this species was the same as in Sadeghian et al. [16]. The small size of the chromosomes could result in undercounting. There is no other report of the chromosome number and the basic chromosome number nor in *Rydingia* neither in *Otostegia* but x = 14 is common among *Leucas* species [9, 10], a closely related genus to *Rydingia* [19].

CONCLUSION

Extensive cytological studies have revealed the presence of polyploid species and diversification in chromosome number in Lamiaceae. Iran, a part of SW Asia is one of the regions of high diversity of this family. This study has been provided the chromosome

number for five species belonging to four genera of Lamiaceae from flora of Iran (*Drepanocaryum*, *Lagochilus*, *Melissa* and *Rydingia*), which may help to promote our understanding of the taxonomic delimitation and the evolutionary events among the taxa of this family.

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