

CHROMOSOME NUMBERS AND KARYOTYPE FEATURES OF EIGHT SPECIES OF APIACEAE IN IRAN

A. Akhavan Roofigar, Z. Khazaei, F. Asadi-Corom & A. Jalili

Received 2023.04.22; accepted for publication 2023.05.31

Akhavan Roofigar, A., Khazaei, Z., Asadi-Corom, F. & Jalili, A. 2023.06.30: Chromosome numbers and karyotype features of eight species of Apiaceae in Iran. *Iran. J. Bot.* 29 (1): 73-77. Tehran.

This study represents chromosomal data for eight species of four genera from Apiaceae. The karyotype analysis in this research was performed using the squash technique on mitotic metaphase chromosomes. Out of the eight species studied, five are new reports (*Dorema aucheri*, *Ducrosia assadii*, *Ferula oopoda*, *F. pseudalliacea*, and *F. sphenobasis*), while three counts confirm (*Dorema ammoniacum*, *Ducrosia anethifolia*, and *Smyrniium cordifolium*) the findings of the previous studies. All species have a basic chromosome number of $x=11$. *Dorema ammoniacum* and *D. aucheri* have only one submetacentric chromosome while in *Ferula pseudalliacea*, about half of the chromosomes were submetacentric. In *Ducrosia anethifolia*, *D. assadii*, *Ferula oopoda*, *F. sphenobasis*, and *Smyrniium cordifolium* all chromosomes are metacentric.

Azadeh Akhavan Roofigar (correspondence <a.akhavan@areeo.ac.ir>), Natural Resources Research section, Isfahan Agricultural and Natural Resources Research and Education Center, AREEO, Isfahan, Iran. -Zahra Khazaei, Department of Plant and Animal Biology, Faculty of Biological Science and Technology, University of Isfahan, Isfahan, Iran. -Fereshteh Asadi-Corom, Biotechnology Research division, Research Institute of Forests and Rangelands, Agricultural Research, Education and Extension Organization, AREEO, Tehran, Iran & Adel Jalili, Botany Research Division, Research Institute of Forests and Rangelands, Agricultural Research, Education and Extension Organization, AREEO, Tehran, Iran.

Keywords: Umbelliferae; chromosome counts; karyotype; idiograms; Iran

اعداد کروموزومی و ویژگی‌های کاریوتایی هشت گونه از تیره چتریان در ایران
آزاده اخوان روفیگر: استادیار پژوهش، بخش تحقیقات منابع طبیعی، مرکز تحقیقات و آموزش کشاورزی و منابع طبیعی استان اصفهان، سازمان تحقیقات، آموزش و ترویج کشاورزی، اصفهان، ایران
زهرا خزائی: دانشجوی دکتری، گروه زیست شناسی گیاهی و جانوری، دانشکده علوم و فن‌آوری‌های زیستی، دانشگاه اصفهان، اصفهان، ایران
فرشته اسدی کرم: محقق، بخش تحقیقات بیوتکنولوژی، مؤسسه تحقیقات جنگلها و مراتع کشور، سازمان تحقیقات، آموزش و ترویج کشاورزی، تهران، ایران
عادل جلیلی: استاد پژوهش، بخش تحقیقات گیاه‌شناسی، مؤسسه تحقیقات جنگلها و مراتع کشور، سازمان تحقیقات، آموزش و ترویج کشاورزی، تهران، ایران
در این مطالعه تحلیل کروموزومی هشت گونه از چهار جنس از تیره چتریان گزارش و مورد بررسی قرار می‌گیرد. از میان گونه‌های مطالعه شده، پنج گونه (*Dorema aucheri*, *Ducrosia assadii*, *Ferula oopoda*, *F. pseudalliacea*, *F. sphenobasis*) برای اولین بار گزارش می‌شوند

و سه گونه (*Dorema ammoniacum*, *Ducrosia anethifolia*, *Smyrniium cordifolium*) تأیید گزارشات قبلی است. عدد پایه کروموزومی برای تمام گونه ها $x=11$ می باشد. گونه های *Dorema ammoniacum* و *D. aucheri* تنها یک کروموزوم متاستریک دارند در حالی که در گونه *D. assadii*, *Ducrosia anethifolia* در گونه های *Ferula pseudalliacea* حدود نیمی از کروموزومها ساب متاستریک شناسایی شدند. در گونه های *Smyrniium cordifolium* و *F. sphenobasis*, *Ferula oopoda* تمام کروموزومها متاستریک بودند.

INTRODUCTION

The Apiaceae is a widely distributed family of flowering plants consisting of ca. 434 genera and 3,780 species worldwide (Cronquist 1981; Pimenov and Leonov 1993). This family is an important group of plants with significant ecological, economic, and medicinal values. The haploid chromosome numbers range from $n=4$ to $n=84$, within the family (Hamal & al. 1986). This variability in chromosome number reflects the diverse evolutionary history of the family. The flora of Iran is home to a rich diversity of plants in the family, with a total of 131 different genera comprising 365 species of which, 118 are endemic to Iran (Mozaffarian 2020).

A number of studies have been conducted on the chromosome number and karyotype analysis of Apiaceae taxa (Goldblatt 1981-1988; Goldblatt & Johnson 1990-2003; Jaberolansar & al. 2010; Mirzadeh Vaghefi & Jalili 2017). Hitherto, only a limited number of species from this family in Iran have been karyologically investigated. According to the IPCN (Index to Plant Chromosome Numbers, www.tropicos.org/Project/IPCN), the chromosome numbers for most species in this family have not yet been reported. This means that there is still a lot of work to be done to fully understand the chromosomal characteristics of the Apiaceae, both in Iran and worldwide.

The primary objectives of our research are to investigate the chromosome number and provide general information on the karyotype characteristics of selected species from the family. The studied species are *Dorema ammoniacum* D. Don, *D. aucheri* Boiss., *Ducrosia anethifolia* (DC.) Boiss., *D. assadii* Alava, *Ferula oopoda* (Boiss. & Buhse) Boiss., *F. pseudalliacea* Rech.f., *F. sphenobasis* C.C. Towns., and *Smyrniium cordifolium* Boiss.

MATERIALS AND METHODS

The seeds of the studied species were provided by the Natural Resources Gen Bank (Research Institute of Forests & Rangelands). The karyotype analysis in this research was performed using the squash technique on mitotic metaphase chromosomes. To prepare the chromosomes, seeds were germinated in Petri dishes at a temperature of 4°C . The root tips were then treated with a 0.01% aqueous solution of colchicine for 2 hours

at room temperature and 14 hours at 4°C . The root tips were then fixed in Carnoy's solution, which is a mixture of glacial acetic acid and ethanol, and stored in 70% ethanol. The materials were hydrolyzed with 1 N HCl for 10 minutes at a temperature of 60°C and stained in 2% aceto-orcein for 3 hours. After staining, the roots were squashed in 45% acetic acid, and the best metaphase plates were photographed using an Olympus (BX40) microscope. The chromosomes in the karyotype were ordered based on their length, and the chromosome pairs were arranged according to Levan's classification (1964). Stebbins's karyotype asymmetry levels were used to describe asymmetry levels (1971). Idiograms were prepared for all species, and the karyotype asymmetry parameters, including total form percentage (TF%) (Huziwara 1962), arm ratio (AR), percentage karyotype asymmetry index (As K%) (Arano 1963), and index of karyotype symmetry (Syi), (Greilhuber & Speta 1976) were evaluated.

RESULTS AND DISCUSSION

The karyo-morphological evidence can be a valuable source of information for the taxonomy of Apiaceae. Currently, the available data on chromosome numbers for Apiaceae species from Iran is limited. We report, for the first time, chromosome numbers for five species, namely *Dorema aucheri*, *Ducrosia assadii*, *Ferula oopoda*, *F. pseudalliacea*, and *F. sphenobasis*. For other three species (*Dorema ammoniacum*, *Ducrosia anethifolia*, and *Smyrniium cordifolium*) were previously documented (Retina and Pimenov 1977; Vassiljeva & al. 1991; Pimenov & Vassilieva 1983; Ghaffari 2020; Cartier 1983; Ghaffari 1987a; Obeidi & al. 2012, 2012a; Al-Eisawi 1989; Jaberolansar & al. 2010). The cytological details of the studied species are presented in Table 1.

Dorema ammoniacum D. Don is an endemic species of Iran and is distributed in Isfahan, Yazd, Sistan, Baluchestan, Semnan, and Tehran Provinces (Mozaffarian 2007).

Specimen examined: Iran, Kerman, Bardsir, 2670 m, 29 37 97 N, 96 04 28 E, Gene bank code: 46711.

A karyological analysis of this species was conducted, revealing a diploid chromosome number of $2n=2x=22$, with ten metacentric and one submetacentric chromosome (as shown in Fig. 1A; Table 1). This finding confirms the findings of previous

studies (Retina and Pimenov 1977; Vassiljeva & al. 1991; Pimenov & Vassilieva 1983; Ghaffari 2020; Ghaffari & al. 2021).

Dorema aucheri Boiss. is an endemic species of Iran and is distributed in Isfahan, Kohgiluyeh va Boyer-Ahmad, Fars, Kerman, and Tehran provinces.

Specimen examined: Iran, Kerman, Sardoeieh, 2935 m, 29 05 37 N, 57 33 05 E, Gene bank code: 46710.

The results of this study showed a diploid chromosome number of $2n=2x=22$ and a karyotypic formula of $10m + 1sm$ (Fig. 1B, Table 1). Interestingly, the S value (46%) was found to be the lowest among the studied species and lower than *Dorema ammoniacum*. It is important to note that this is the first reported chromosome count for *D. aucheri*, providing new insights into the cytology of this species.

Ducrosia anethifolia (DC.) Boiss. has a distribution range that includes Iraq, Afghanistan, Pakistan, Syria, Arabia, and Iran.

Specimen examined: Iran, Hormozgan, Bandar Abbas, 750 m, 56 11 10 N, 27 49 49 E, Gene bank code: 47168.

Our analysis of samples from this species revealed a chromosome count of $2n=2x=22$ (Fig. 1C), which is consistent with previous reports on the somatic chromosome number by Cartier (1983), Ghaffari (1987a), and Obeidi & al. (2012, 2012a) and Al-Eisawi (1989). The karyotype of this species was composed of

metacentric (11m) chromosomes (Fig. 1C). Compared to other species studied, the chromosomes in this species were found to be shortest in length (Table 1).

Ducrosia assadii Alava is an endemic species found exclusively in Kerman, Iran.

Specimen examined: Iran, Kerman, Rain, 2578 m, 57 22 28 N, 29 17 07 E, Gene bank code: 47220.

This species is diploid, with a chromosome count of $2n=2x=22$. The karyotype of this species was observed to be composed of metacentric chromosomes, with a karyotype formula of $10m+1M$ (Fig. 1D, Table 1). This is the first report of the chromosome number and karyotype details of this species.

Ferula oopoda (Boiss. & Buhse) Boiss. a plant species found in Iraq, Caucasus, and Iran (Mozaffarian 2007), had its chromosome number reported for the first time in this study.

Specimen examined: Iran, Semnan, Damghan, 2300 m, 54 08 23 N, 35 44 29 E, Gene bank code: 47262.

The species was observed to be diploid with a chromosome count of $2n=2x=22$ (Fig. 1E), and its karyotypic formula was found to be $11m$ (Fig. 1E).

Ferula pseudalliacea Rech.f. an endemic species of Iran found in Kurdistan, Kermanshah, and Lorestan (Mozaffarian 2007).

Specimen examined: Iran, Kurdistan, Sanandaj, 1850 m, Gene bank code: 45460

Table 1. Chromosome counts and karyomorphological parameters of the eight examined Apiaceae species. CL, chromosome length; q, long arm; p, short arm; TF%, total form percentage; S%, symmetry index; Ask%, Arano index of karyotype asymmetry; Syi%, index of karyotype symmetry; SC: symmetry classes of Stebbins; KF, karyotype formula.

Species	chromosome number	CL	P	q	TF%	S%	Ask%	Syi%	SC	KF
<i>Dorema ammoniacum</i>	$2n=2x=22$	3.47	1.57	1.9	0.45	0.67	0.54	0.82	1A	$10m+1sm$
<i>Dorema aucheri</i>	$2n=2x=22$	3.1	1.32	1.78	0.42	0.46	0.57	0.73	1B	$10m+1sm$
<i>Ducrosia anethifolia</i>	$2n=2x=22$	2.98	1.31	1.67	0.43	0.58	0.55	0.78	1A	11m
<i>Ducrosia assadii</i>	$2n=2x=22$	4.05	1.88	2.17	0.46	0.46	0.53	0.86	1B	$10m+1M$
<i>Ferula oopoda</i>	$2n=2x=22$	3.36	1.55	1.81	0.46	0.48	0.53	0.85	1B	11m
<i>Ferula pseudalliacea</i>	$2n=2x=22$	3.46	1.24	2.21	0.35	0.6	0.64	0.56	2A	$5m+6sm$
<i>Ferula sphenobasis</i>	$2n=2x=22$	3.14	1.34	1.8	0.42	0.48	0.57	0.74	1B	11m
<i>Smyrniium cordifolium</i>	$2n=2x=22$	3.83	1.64	2.18	0.42	0.56	0.57	0.75	1A	11m

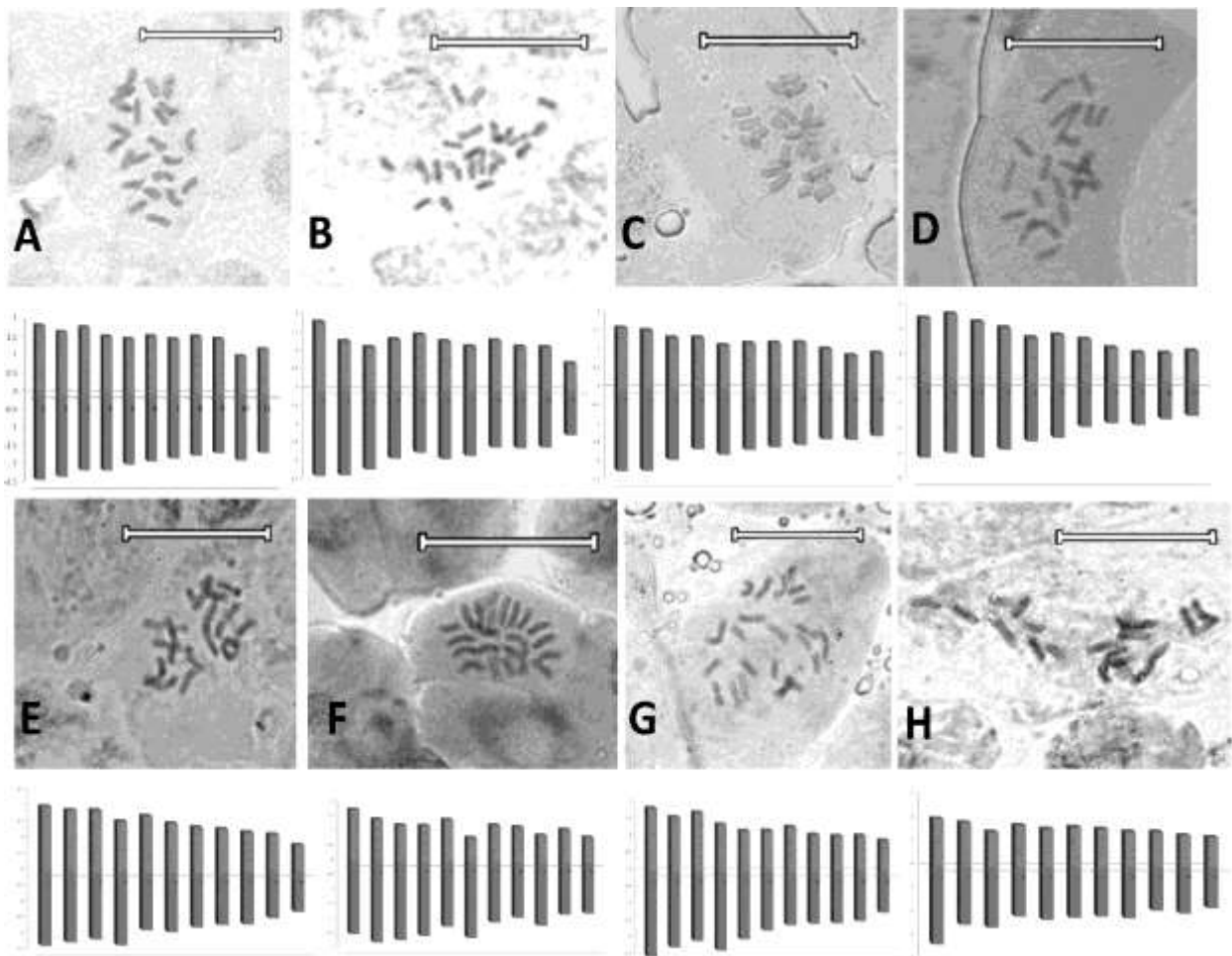


Fig 1. Somatic metaphases and idiograms in A, *Dorema ammoniacum* ($2n=2x=22$); B, *Dorema aucheri* ($2n=2x=22$); C, *Ducrosia anethifolia* ($2n=2x=22$); D, *Ducrosia assadii* ($2n=2x=22$); E, *Ferula oopoda* ($2n=2x=22$); F, *Ferula pseudalliacea* ($2n=2x=22$); G, *Ferula sphenobasis* ($2n=2x=22$); H, *Smyrniium cordifolium* ($2n=2x=22$). Scale bar 20 μm .

This species was observed to be diploid with a chromosome count of $2n=2x=20$. Of the total chromosome pairs, five pairs in our sample were found to be metacentric (m) and the remaining six pairs were submetacentric (sm) (Fig. 1F).

Ferula sphenobasis C.C. Towns. is a species reported to be found in Iran and Iraq.

Specimen examined: Iran, Khuzestan, Masjed Soleyman, 554 m, 35 45 93 N, 34 50 68 E, Gene bank code: 46530.

Our analysis showed that this species is diploid with a chromosome count of $2n=2x=22$. The karyotype of this species was observed to consist of 11 pairs of metacentric (m) chromosomes (Fig. 1G).

Smyrniium cordifolium Boiss. is a plant species that grows in Turkey, Afghanistan, various regions of central Asia, and most parts of Iran.

Specimen examined: Iran, Chaharmahal va Bakhtiari, Lordegan, 2862 m, 51 15 39 N, 31 26 30 E, Gene bank code: 46316.

Our analysis of a sample from this species revealed it to be diploid with a chromosome count of $2n=22$ (Fig. 1H). This count is consistent with previous reports by Montmollin (1986), Queiros (1974), Vachova (1976), and Strid (1983) for different species of *Smyrniium*, and Jaberalansar & al. (2010) for *S. cordifolium* on samples collected from different parts of Iran.

ACKNOWLEDGMENTS

The authors are thankful to the Natural Resources Research Section of Isfahan Agricultural and Natural Resources Research and Education Center, (AREEO), Isfahan, and also the Research Institute of Forests and Rangelands of Iran (RIFR) for supporting this study.

REFERENCES

- Al-Eisawi, D.M. 1989: Chromosome counts of Umbelliferae of Jordan. -Annali di Botanica 47: 201–214.
- Arano, H. 1963: Cytological studies in subfamily Carduoideae (Compositae) of Japan. IX. The karyotype analysis and phylogenetic considerations on *Pertya* and *Ainsliaea*. -Botanical Magazine, Tokyo 76: 32–39.
- Cronquist, A. 1981: An integrated system of classification of flowering plants. -Columbia University Press, Columbia.
- Ghaffari, S.M. 1987a: Chromosome counts of some angiosperms from Iran. -Iranian Journal of Botany 3(2): 183–188.
- Ghaffari, S.M. 2020. Index to Plant Chromosome Number of Iran, 1st ed. -Research Institute of Forests and Rangelands. Tehran, Iran; 336p.
- Ghaffari, S.M., Ghamari Zare, A., Asadi Corom, F. & Sedaghati, M. 2021: Chromosome number and meiotic behavior in several plant taxa from Iran. -Botanica Serbica 45(2): 333–339.
- Goldblatt, P. 1981, 1984, 1985, 1988: Index to plant chromosome numbers. 1975-1978, 1979-1981, 1982-1983, 1984-1985. -Monographs in Systematic Botany from the Missouri Botanical Garden 5, 8, 13, 23.
- Goldblatt, P. & Johnson D.E. 1990, 1991, 1994, 1996, 1998, 2000, 2003: Index to plant chromosome numbers. 1986-1987, 1988-1989, 1990-1991, 1992-1993, 1994-1995, 1996-1997, 1998-2000. - Monographs in Systematic Botany from the Missouri Botanical Garden 30, 40, 51, 58, 81, 94.
- Greilhuber, J. & Speta, F. 1976: C-banded karyotypes in the *Scilla hohenackeri* group, *S. persica* and *Puschkinia* (Liliaceae). -Plant Systematics & Evolution 126: 149–188.
- Hamal, I.A., Langer, A. & Koul, A.K. 1986: Nucleolar organizing region in the Apiaceae (Umbelliferae). - Plant Systematics & Evolution 154: 11–30.
- Huziwara, Y. 1962: Karyotype analysis in some genera of Compositae. VIII. Further studies on the chromosomes of Aster. -American Journal of Botany. 49: 116–119.
- Levan A, Fredgra K, Sandberg A.A. 1964: Nomenclature for centromeric position on chromosomes. -Hereditas 52: 201–220.
- Mirzadeh Vaghefi, S.S. & Jalili, A. 2017: Chromosome Counts of some Iranian plants. -Iranian Journal of Botany 23 (2):136–139.
- Montmollin, B.d. 1986: etude cytotonomique de la flore de la Crète. III. Nombres chromosomiques. - Candollea 41: 431-439.
- Mozaffarian V. 2007: Umbelliferae. In: Flora of Iran (eds. Assadi, M., Khatamsaz, M., Maassoumi, A.A.) No. 54. -Tehran (in Persian).
- Mozaffarian, V. 2020: A short survey of the plants of the Umbelliferae (Apiaceae) family in Iran and their value and importance (Monospecific genus, geographical distribution, endemism, medicinal, and other uses). -Iran Nature 5 (24): 43–67.
- Pimenov, M.G. & Leonov, M.V. 1993: Genera of the Umbelliferae, Royal Botanic Gardens Kew Press, London.
- Pimenov, M.G. & Vassiljeva, M.G. 1983: In IOPB chromosome number reports LXXXI. -Taxon 32: 663-664.
- Queiros, M. 1974: Contribucaopara o conhecimento citotaxonomico das Spermatophyta de Portugal. VII. Umbelliferae, Suppl. 1.- Bol. Soc. Brot., sér. 2 48: 171–186.
- Retina, T.A. & Pimenov, M.G. 1977: Chromosome numbers in species of the family Umbelliferae of middle Asia. -Biol. Nauki (Moscow) 7: 90–95.
- Stebbins, G. L. 1971: Chromosome evolution in higher plants. -Edward Arnold Publisher, London.
- Strid, A. 1983: In IOPB chromosome number reports LXXVIII. -Taxon 32: 138–140.
- Vachova, M. 1976: In Index of chromosome numbers of Slovakian flora. Part 5. -Acta Fac. Rerum Nat. Univ. Comeniana, Bot. 25: 1–18.
- Vassiljeva, M.G., Alexeeva, T.V. Pimenov M.G. & Kljuykov, E.V. 1991: IOPB chromosome data 3.- International Organization of Plant Biosystematists Newsletter (Zurich) 17: 10–13.