IRANIAN JOURNAL OF BOTANY 27 (1), 2021
DOI: 10.22092/ijb.2021.353009.1306

CHROMOSOME COUNTS FOR NINE SPECIES OF ACANTHOPHYLLUM (CARYOPHYLLACEAE) FROM IRAN

S. M. Ghaffari

Received 2020. 12. 24; accepted for publication 2021. 04. 07

Ghaffari S. M. 2021. 06. 30: Chromosome counts for nine species of *Acanthophyllum (*Caryophyllaceae) from Iran. - *Iran. J. Bot. 27 (1): 53-57.* Tehran.

Original meiotic or mitotic chromosome numbers are reported for the nine species of *Acanthophyllum*, viz: *A. acerosum* Sosn. (n=28), *A. crassinodum* Yukhan. & Edmondson (2n=90), *A. elatius* Bunge (n=15+1B, 2n=30+1B), *A. glandolusum* Bunge ex Boiss. (n=90), *A. gracile* Bunge ex Boiss. (2n=30), *A. korshinskyi* Schischk. (2n=30), *A. microcephalum* Boiss. (2n=60), *A. pachycephalum* Schiman - Czeika (2n=30), *A. sordidum* Bunge ex Boiss. (2n=60). Mitotic counts for all species (except *A. microcephalum* and *A. glandolusum*), meiotic count for *A. acerosum* and presence of B chromosomes in *A. elatius* is reported here for the first time.

Seyed Mahmood Ghaffari (correspondence<ghaffari@ibb.ut.ac.ir>) Institute of Biochemistry and Biophysics, University of Tehran, Tehran, Iran.

Key words: *Acanthophyllum*; Caryophyllaceae; chromosome count; Iran

شمارش کروموزومی ۹ گونه چوبک (Acanthophyllum), (Caryophyllaceae) از ایران

سید محمود غفاری: استاد مؤسسه تحقیقات بیوشیمی و بیوفیزیک دانشگاه تهران، تهران، ایران

شمارش کروموزومی ۹ گونه چوبک (Acanthophyllum) شامل: A. acerosum Sosn. (n=28); A. crassinodum Yukhan. & Edmondson (2n=90); A. elatius Bunge (n=15+1B, 2n=30+1B); A. glandolusum Bunge ex Boiss. (n=90); A. gracile Bunge ex Boiss. (2n=30); A. korshinskyi Schischk. (2n=30); A. microcephalum Boiss. (2n=60); A. pachycephalum Schiman - Czeika (2n=30); A. sordidum Bunge ex Boiss. (2n=60) گزارش می‌شود. شمارش کروموزومی میتوزی برای تمام گونه‌ها بجز (A. microcephalum و A. glandolusum)، شمارش کروموزومی میوزی برای گونه A. acerosum و حضور B کروموزوم برای گونه A. elatius برای اولین بار گزارش می‌شود.

INTRODUCTION

Meyer (1831) introduced the genus *Acanthophyllum* by the species *Acanthophyllum mucronatum* C. A. Mey. for the first time. These plants are normally found in the dry areas, deserts, slopes of mountains, as well as temperate zones. According to the floristic regions of Takhtajan (1986), *Acanthophyllum* belongs to the Irano-Turanian region, extending from Pamiro-Alajian, Tienshan, N xinjiang (Kazakhstan, Mongolia) in the north-east to Syria in the West. *Acanthophyllum* with a total of c. 75 species of which 52 are endemics to the area, is remarkable in various respects. Schiman-Czeika (1988) recognized 4 sections including 33 species in Iran. Later, two new *Acanthphyllum* species (*A. ejtehadii* Mahmoudi & Vaezi and *A. yasammin-nassehiae* Joharchi & Pirani) were described by Mahmoudi Shamsabad & al. (2012) and Pirani & al. (2013) respectively. Joharchi & Akhani (2006), reported *A. kandaharicum* Gilli and *A. stenostegium* Freyn from Nehbandah, neighboring Pakistan and Kalat-e-Naderi in Khorassan province. *Acanthophyllum maimanense* Rech. F. & Schiman-Czeika was recorded from Torbate Jam in Khorasan province by Mahmoudi Shamsabad & al. (2012). The total numbers of *Acanthophyllum* species in the flora of Iran, now are 38 which belong to four sections that is about 51.35% of those known for the world.

According to the previous chromosome counts

(Nussbaumer 1964, Aryavand & Favarger 1980, Ghaffari 1986, 1987, 1988, 2002, 2004, Turner 1994, Maleki Sadabadi & al. 2017), the basic chromosome numbers for the genus *Acanthophyllum* are x =14 and x =15. In this paper meiotic or mitotic chromosome counts for nine species are presented.

MATERIALS AND METHODS

For mitotic analysis actively growing root tips were used. Roots were pretreated with 0.002 M 8-hydroxyquinoline at 20°C for 3 hr, and then fixed in 3:1 (ethanol: glacial acetic acid). Staining was carried out with the Feulgen reaction enhanced by squashing in 2% acetocarmine. For meiotic studies floral buds of appropriate size were fixed in the Piennar′s fixing fluid (ethanol 96%, chloroform, propionic acid, 6:3:2 V/V). Staining was carried out with acetocarmine. One sample of each species were sent to Wein Herbarium to be identified by Schiman-Czeikam.

RESULTS

Meiotic or mitotic chromosome counts for 9 species are presented as follow:

Section *Acanthophyllum*

Acanthophyllum acerosum Sosn.

Qazvin: Avaj, 16 Km. towards Hamadan, 1,100 m, Ghaffari 176.

Distribution: East of Turkey, NE of Iraq, NW of Iran.

Meiosis in this taxon was regular and showed 28 bivalents at diakinesis and metaphase I (fig,1A). At metaphase II 28 chromatids in each pool were observed (fig. 1B). The chromosome number for this species is given here for the first time. As indicated in my previous paper (Ghaffari 2004), basic chromosome number in the genus *Acanthophyllum* is x =15, except *A. caespitosum (*of the section *Oligosperma*) which is a diploid species with n = 14 (x = 14). So, *A. acerosum* is the second species which represents x=14 in the genus *Acanthophyllum*. In the recent years Pirani & al. (2014, 2020), by a phylogeny analysis of molecular data, placed the *A. caespitosum* and *A. acerosum in* the section *Acanthophyllum*. It seems that this revision by molecular phylogeny is close to reality. *Acanthophyllum acerosum* is a tetraploid species that represents x=14 and has some morphological and cytological similarity with *A. caespitosum*. Thus, the section *Acanthophyllum* has two basic numbers including x=14 and 15, but section *Oligosperma* has only one basic number of x= 15.

A. microcephalum Boiss.

Tehran: Jajroud, Ghaffari 16664

Distribution: Many parts of Iran, South of Turkmenistan.

This taxon is tetraploid with chromosome of 2n = 60, which most of them were metacentric (fig. 1C). Previous gametic number of n = 30 is reported for this species by Aryavand & Favarger (1980) and Ghaffari (2004) from different parts of Iran. Also, mitotic count (2n=60) was reported by Nussbamer (1964) from Turkmenistan. This mitotic number is reported here for the first time from a new locality in Iran.

Section *Macrostegia* Boiss.

A. gracile Bunge ex Boiss.

Khorasan Razavi: Mashhad towards Torbat e-Heydariyeh. Ghaffari 5765

Distribution: North and NE of Iran, Turkmenistan.

Only one previous gametic number for this species exists, n = 15 by Ghaffari (2004), which is in agreement with the present count of 2n = 30 (fig.1D). Mitotic count for this species is reported here for the first time. Basiri- Esfahani & al. (2011) reduced this distinct species to the rank of variety (*A. bracteatum* var. *gracile*), which is very questionable. Pirani & al. (2014, 2020) by molecular phylogeny confirmed the independence of this species.

A. pachycephalum Schiman - Czeika

Karaj: 5 Km towards Chalus. Ghaffari 14164

Distribution: Endemic to western and central parts of Iran.

This diploid species has a symmetrical karyotype with fifteen pairs of metacentric or submetacentric chromosomes (2n=30) which one pair having a satellite on the short arm (fig.1E). Only one gametic number of n = 15 is reported for this species by Ghaffari (2004).

Basiri-Esfahani & al. (2011) reduced the rank of this taxon to variety (*A. bracteatum* var. *pachycephalum*), which is not approved by Pirani & al. (2014, 2020).

Section *Oligosperma* Schischk.

A. elatius Bunge

Khorasan Razavi: Sabzevar towards Abbasabad. Ghaffari 7366.

Distribution: East of Iran, Tajikistan, northwest of Afghanistan.

Meiotic study showed 15 bivalents at metaphase I, which is in agreement with the previous report by Ghaffari (2004). In some cells one B chromosome were observed (fig. 1F). Somatic chromosome counts of 25 root tips disclosed a chromosome number of 2n = 30 + 0-1 B chromosome which is confirmed the meiotic results (fig.1G, H). The karyotype consisted of eight metacentric pairs, four submetacentric pairs and three

pairs of acrocentric chromosomes. One pair of submetacentric chromosome possessed a satellite in its short arm. Mitotic count and presence of B chromosome in this species are reported here for the first time.

A. korshinskyi Schischk.

Khorasan Razavi: between Sabzevar and Mehr, Ghaffari 3165

Distribution: Northeast of Iran, west of Afghanistan, Turkmenistan.

Previous chromosome count on meiotic division (n = 15) for this species was reported by Ghaffari (2004). Karyotype in this species includes 15 pairs of metacentric and submetacentric chromosomes (fig.1I). This mitotic count is reported here for the first time.

Section *Pleiosperma* Boiss.

A. crassinodum Yukhan. & Edmondson

North Khorasan: Darreh Gaz, Tandureh National park, Ghaffari 2666

Distribution: Endemic to Iran, Afghanistan and Turkmenistan.

This species is hexaploid with 2n = 6x = 90 (fig.1J), which is in agreement with the previous report on gametic number of n = 45 by Ghaffari (2004). As indicated in my previous paper (Ghaffari 2004), this species is very close to *A. glandolusum* from morphological and cytological characteristics point of view. For this reason, Basiri Esfahani & al. (2011) regarded this species as a synonym of *A. glandolusum*.

A. glandolusum Bunge ex Boiss.

Khorasan Razavi: Ghochan towards Darreh Gaz. Ghaffari 17164

Distribution: Many parts of Iran, SW of Afghanistan, Turkmenistan, Tajikistan.

This taxon is hexaploid and showed chromosome compliment of 2n = 90 (fig.1K). The previous report on gametic number (n=45) for this species is reported by (Ghaffari 2004). Also, mitotic count (2n=90) has been reported by Nussbaumer (1964) from Turkmenistan. This mitotic count is reported here for the first time for flora of Iran.

A. sordidum Bunge ex Boiss.

Tehran: Garmsar, Behbar, Ghaffari 5866

Distribution: Iran, Afghanistan, Pakistan and Turkmenistan.

This species is tetraploid and showed 60 chromosomes at metaphase of mitosis (fig.1L). All chromosomes were small and they were so similar to the chromosomes of *A. glandolusum* and *A. crassinodum* from the same section. This count in agreement with previous report on meiotic study (n = 30) by Ghaffari (2004). By the results of molecular phylogeny, Pirani & al. (2014, 2020) believed that *A. sordidum* should be removed form section *Pleiosperma.*

DISCUSSION

According to the present work and previous reports, the basic chromosome number for the genus *Acanthophyllum* are x=14 and 15. Basic chromosome number of x=14 and 15 belongs to the section *Acanthophyllum* which has two races of diploidy (2n=2x=30) and tetraploidy (2n=4x=60). At this time, section *Oligosperma* has only one basic chromosome number of x=15 with high number of species in the genus of *Acanthophyllum.* All species which are considered in sections *Oligosperma* and *Macrostegia* are diploid with 2n=2x =30. The members of section *Pleiosperma* are hexaploid with 2n=6x=90 (except *A. sordidum* with 2n=60). Pirani & al. (2014) believed that *A. sordidum* should be excluded from section *Pleiosperma* which is questionable. Because this taxon in morphological and cytological point of view is similar to the members of section *Pleiosperma*. From the 3 species of the section *Paniculata*, there is only one report for *A. gypsophiloides* (2n=30) by Nussbaumer (1964) from Uzbekistan (Tashkent) so far. Unfortunately, for the other sections there are no chromosome number reports. It seems that for accurate classification of the genus *Acanthophyllum*, the morphological, anatomical, cytological and molecular characters should be investigated and considered.

ACKNOWLEDGEMENT

This work was supported by Research council, University of Tehran. I would like to thank Dr. Schiman-Czeika for her help in the identification of the plants.

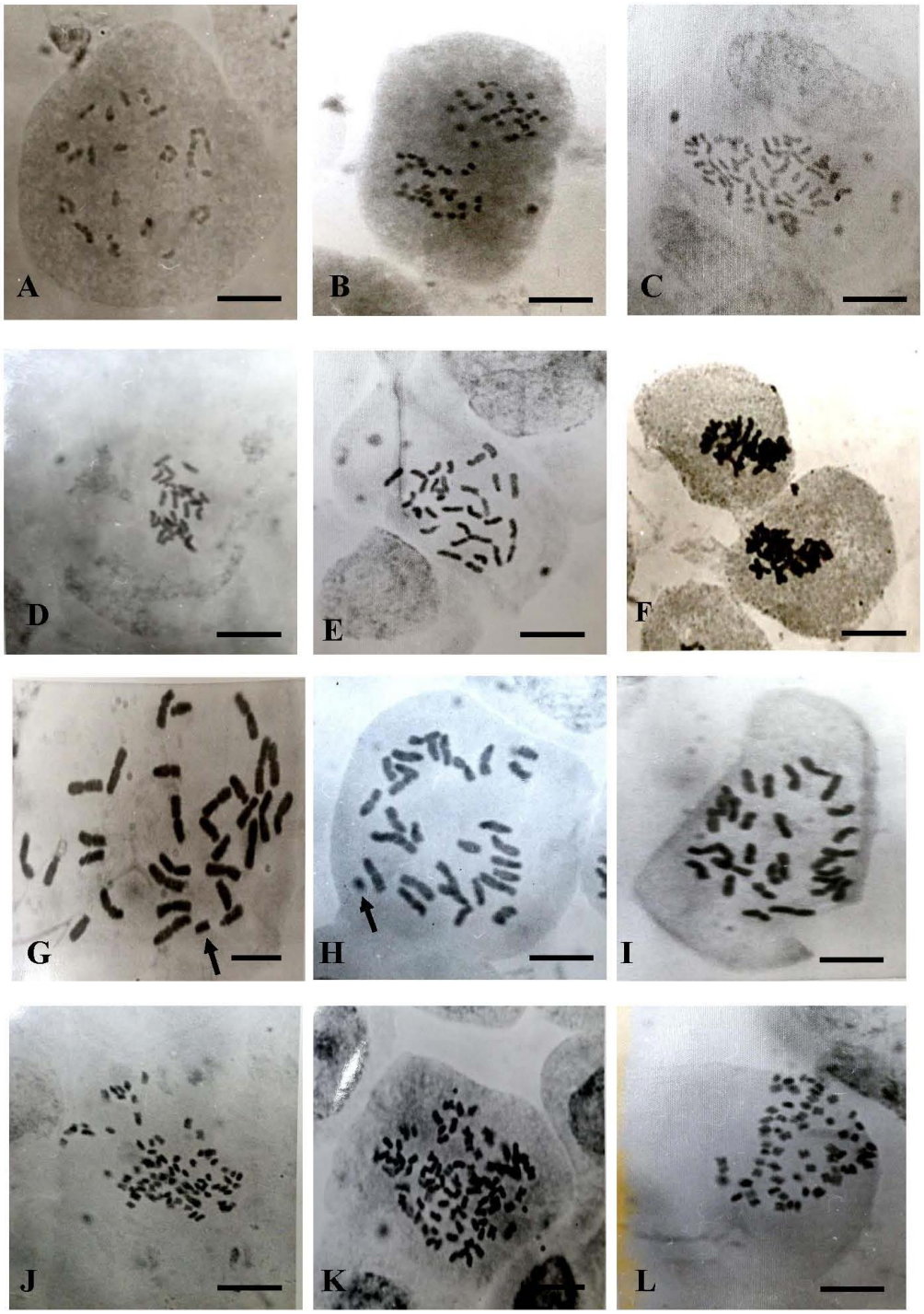

Fig. 1. Meiosis & mitosis photomicrograph of *Acanthophyllum* species. A & B, *Acanthophyllum acerosum*, A, diakinesis (n=28), B, anaphase I, showing (18-18) chromosome segregation; C, *A. microcephalum*, metaphase (2n=60); D, *A. gracile*, metaphase (2n=30); E, *A. pachycephalum*, metaphase (2n=30); F, G, H, *A. elatius*, F, metaphase I, showing B chromosome in the corner of bottom cell; G & H, metaphase, showing 2n=30+1B (arrow); I, *A. korshinskyi*, metaphase (2n=30); J, *A. carssinodum*, metaphase (2n= 90); K, *A. glandolusum*, metaphase (2n=90); L, *A. sordidum*, metaphase (2n=60). Scale bar 5 um.

REFERENCES

Aryavand, A & Favarger, C. 1980: Contribution a l' etude cytotaxonomique des Caryophyllacees de l Iran. -Rev. Biologie. Ecol. Mediterra. 3/1: 15-26.

Basiri Esfahani, SH., Bidi, B., Assadi, M. & Rahimi Nejad, M. R. 2011: A taxonomic study of *Acanthophyllum* C. A. Mey. (Caryophyllaceae) in Iran. -Iran. J. Bot. 17 (1): 24-39.

Ghaffari, S. M. 1986: Chromosome number reports XCIII. -Taxon. 35/4: 901.

Ghaffari, S. M. 1987: Chromosome number reports CVI. -Taxon. 36/3: 659.

Ghaffari, S. M. 1988: Chromosome number reports XCIX. -Taxon. 37/2: 397.

Ghaffari, S. M. (1998): Chromosome studies and distribution of nine species of *Cousinia* section *Stenocephalae* (*Asteraceae*) in Iran. -Bull. Soci. Neuch. Sci. 121: 61-68.

Ghaffari, S. M. 2002: Presence and behavior of B-chromosomes in *Acanthophyllum laxiusculum (Caryophyllaceae*). -Genetica 115: 319-323.

Ghaffari, S. M. 2004: Cytotaxonomy of some species of *Acanthophyllum* (*Caryophyllaceae*) from Iran. -Biologia 59 /1: 53 - 60. Joharchi, M. R. & Akhani, H. 2006: Notes on the flora of Iran 6: eight new plant records from Iran collected from Khorasan and Golestn provinces (N. E. Iran). -Rostaniha 7 (2): 131-141.

Mahmoudi Shamsabad, M., Vaezi, J., Memariani F. & Joharchi, M. R. 2012: A new species and a new record of *Acanthophyllum* C. A. Mey. (Caryophyllaceae) from northeast of Iran. - Iran. J. Bot. 18 (1): 59-63.

Maleki Sadabadi, Z., Ejtehadi, H., Abrishamchi, P., Vaezi J., & Erfanian Taleii Noghan, B.M. 2017: Comparative study of autecological, morphological, anatomical and karyological characteristics of *Acanthophyllum ejtehadii* Mahmoudi & Vaezi (Caryophyllaceae): a rare endemic in Iran. -Taiwania 62 (3): 321-330.

Meyer, C. A. 1831: *Verzeichniss der pflanzen Caucasas*. Petersburg, pp. 210.

Nussbaumer, F. 1964: Nombers chromosomiques nouveaux ches les Caryophyllaceae.-Bull. Soc. Neuch. Sci. nat. 57: 171-180.

Pirani, A., Joharchi, M. R. & Memariani, F. 2013: A new species of *Acanthophyllum (*Caryophyllaceae) from Iran. -Phytotaxa 92 (1): 20-24.

Pirani, A., Zarre, S., Pfeil, B.E., Bertrand, Y. J. K., Assadi, M. & Oxelman, B. 2014: Molecular phylogeny of *Acanthophyllum (*Caryophyllaceae, Caryophylleae) with emphasis on infarageneric classification. -Taxon 63: 592-607.

Pirani, A., Moazzeni, H., Zarre, S., Rabeler, R. K., Oxelman, B., Pavlenko, A.V. & Kovalehuk, A. 2020: Phylogeny of *Acanthophyllum* s. l. revisited: An update on generic concept and sectional classification. -Taxon published online.

Schiman-Czeika, H. 1988: *Acanthophyllum*. In: K. H. RECHINGER (ed.), *Flora Iranica*. 163, Graz, pp. 253-329.

Takhtajan, A. L. & A. A. Fedorov, A. A. 1972: Flora of Yerevan. Leningerad, 394p (in Russian).

Turner, B. L. 1994: Chromosome numbers and their phyletic interpretation. In: Behnke H. D., Mabry T. J. (eds) Caryophyllales. Springer, Berlin. Pp 27-43.