Journal of Applied Biological Sciences 5 (3): 21-25, 2011
ISSN: 1307-1130, E-ISSN: 2146-0108, www.nobel.gen.tr

Karyotypic Study of Some Salvia Lamiaceae Species from Iran

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Received: May 21, 2011
Accepted: May 25, 2011

Abstract

The chromosome numbers of eight Salvia L. species is reported. The taxa revealed the chromosome numbers varied between 2n = 14, 16, 20 and 22. The basic chromosome numbers of the studied species were x= 7,8,10 and 11. The ploidy level of all species was diploid and the chromosome numbers of four species are reported for the first time in Iran. The karyotype details of eight species mostly show the median point, median region, submedian region, subterminal region, terminal region, and terminal point chromosome centromeres which are reported for the first time in Iran.

Keywords: Chromosome numbers, Ideogram, Salvia, ploidy level

INTRODUCTION

The genus Salvia L. has over 900 species all around the world including about 55 species in Iran. It is distributed in subtropical, temperate, sub- arctic and arctic areas as well as in the tropical regions of Iran [1]. Some of these species are perennial, herbaceous, suffruticose, fruticose and subshrubby [1]. This genus is quite well- known for its medical properties [2].

Studies on the chromosome of this genus are difficult because the sizes of their chromosomes are too small [3]. Chromosome numbers have been reported for some of the Salvia species [4,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19, 20]. In addition, Ozdemir and Senel [13] reported karylogical properties of S. sclerea L. in Turkey. Ozkan and Soy [18] displayed the karyotype structure of S. blepharoclaena Hedge & Hubb.-Mor. from Turkey.

The chromosome numbers of the various species of Salvia in Iran are unknown. Since the Salvia species are distributed in this region, the aim of the present study is to provide chromosomal data for this gene pool of the Salvia genus. The species studied are S. spinosa L., S. macrosiphon Boiss., S. atropatana L., S. sharifii Rech. f. & Esfand., S. sclarea L., S. nemorosa L. ssp. tesquicola, S. virgata Jacq. and S. ceratophylla L. Some chromosome counts and all the karyotype patterns have been conducted for the first time in Iran. Some of the counts confirm those contained in previous reports and some are different.

MATERIALS AND METHODS

Plant material: 23 accessions of eight Salvia species collected from natural habitats in Iran are presented (Table 1).

Chromosome study: For mitotic studies, the seeds collected from various accessions were germinated in sterilized Petri dishes. Then root tips were pre-treated with an ice bath at 4º C for 18 hours and then fixed in a mixture of ethanol: acetic acid (3:1, respectively) for 24 hours. The root tips were macerated in a 1N HCl solution at 60º C for about 5 minutes. A squash technique was used for cytological studies with 2% aceto-orcein solution [3]. The clearest mitotic metaphase among 25 cells was photographed using an OLYMPUS BX50 photomicroscope. Ideograms prepared from mitotic metaphase. Chromosome measurements (Total Form percentage and Total haploid chromosome length) were based on five metaphase plates [18]. Voucher specimens of the taxa studied were deposited in the Herbarium of Shahrekord University.

RESULTS AND DISCUSSION

S.Spinosa

The results of this study showed that the chromosome number of S. spinosa is 2n=20 and this is reported for the first time in Iran (Figure 1A). Based on karyotypic study, the 1st, 3rd, 4th, 5th, 6th and 10th chromosomes have subterminal region (st) centromeres, 2nd, 7th and 9th chromosomes have terminal region (t) centromeres and 8th chromosome has terminal point (T) centromere. The chromosome lengths ranged from 0.48 to 2.25 µm (Figure 2a, Table 2)

S. Macrosiphon

Cytological studies revealed that the chromosome number of S. macrosiphon is 2n=20 (Figure 1B). The 1st, 3rd, 4th, 8th, 9th and 10th chromosomes have terminal point (T) centromeres, 2nd chromosome has median region (m) centromere, 7th chromosome has median point (M) centromere, and 5th and 6th chromosomes have subterminal region (st) centromeres. The chromosome lengths ranged from 0.4 to 1.9 µm (Figure 2b, Table 2).

S. Atropatana

The chromosome number of S. atropatana is 2n=20 (Figure 1C). This is also the first time that the chromosome number of this species has been reported for Iran. This taxon is considered an Irano-Turanian element with high variations in morphological characters [1,21]. The karyotype of this species displayed that the 1st, 2nd, 6th and 10th chromosomes have terminal point (T) centromeres, 3rd chromosome has submediam region (sm) centromer, 5th and 9th chromosomes have terminal region (t) centromeres, 4th chromosome has median point (M) centromere, and 7th and 8th chromosomes have subterminal region (st) centromeres. The chromosome lengths were in the range of 0.25 to 0.9 µm (Figure 2c, Table 2).

S. Sharifii

Cytological studies revealed that the chromosome number of S. sharifii is 2n=20 (Figure 1D). The chromosome count of this Iranian endemic species is recorded here for the first time. The 1st chromosome has terminal region (t) centromere, 2nd and 6th chromosomes have subterminal region (st) centromeres, 3rd and 4th chromosomes have submedian region (sm) centromeres, 5th and 8th chromosomes have median point (M) centromeres, and 7th, 9th and 10th chromosomes have terminal point (T) centromeres. The chromosome lengths ranged from 0.35 to 0.98 µm (Figure 2.d, Table 2).

S. Sclarea

The chromosome number of S. sclarea is 2n= 22 (Figure 1E). Based on karyotypic study, the 1st, 7th and 8th chromosomes have terminal region (t) centromeres, 2nd chromosome has median region (m) centromere, 3rd, 4th and 6th chromosomes have subterminal region (st) centromeres, 5th and 9th chromosomes have median point (M) centromeres, 10th chromosome has terminal point (T) centromere and 11th chromosome has submedian region (sm) centromere. The chromosome lengths were in the range of 0.3 to 0.7 µm (Figure 2.e, Table 2).

S. Nemorosa Ssp. Tesquicola

The chromosome number of S. nemorosa ssp. tesquicola is 2n= 14 (Figure 1F). The karyotype of this species showed that the 1st chromosome has terminal region (t) centromere, 2nd chromosome has submedian region (sm) centromere, 3rd, 5th and 6th chromosomes have terminal point (T) centromeres, and 4th and 7th chromosome have median region (m) centromeres. The chromosome lengths were in the range of 0.38 to 1.5 µm (Figure 2.f, Table2).

S. Virgata

The chromosome number of S. virgata was 2n= 16 (Figure 1G). The 1st chromosome has terminal region (t) centromere, 2nd, 3rd, 6th, 7th and 8th chromosomes have terminal point (T) centromeres, 4th chromosome has subterminal region (st) centromere and 5th chromosome has submedian region (sm) centromere. The chromosome lengths were in the range of 0.3 to1.3 µm (Figure 2.g, Table 2).

S. Ceratophylla

The chromosome number of S. ceratophylla is 2n= 14 (Figure 1H). The chromosome number of this species is first reported for Iran. The 1st, 3rd, 4th and 7th chromosomes have terminal region (t) centromeres, 2nd and 5th chromosomes have submedian region (sm) centromeres, and 6th chromosome has terminal point (T) centromere. The chromosome lengths were in the range of 0.25 to1.1 µm (Figure 2.h, Table 2).

According to the literature, some of Salvia species are characterized by several basic chromosome numbers such as x= 6 [5,6], x=7, 8, 9 [5,10], x=10, 11, 13, 15, 16, 17, 19 [4, 5,7,9,10,15,16,22,23], and x=44 [6], implying that Salvia has more than one basic chromosome number. Based on the basic chromosome number reported above, it appears that the basic numbers are x= 7, 8, 10, 11 which are observed in S. macrosiphon (x=10), S. atropatana (x=10), S. spinosa (x=10), S. sharifii (x=10), S. nemorosa ssp. tesquicola (x=7), S. virgata (x=8), S. ceratophylla (x= 7) and S. sclarea (x= 11). Consequently, these Salvia species are thought to be diploid which is in accordance with the literature. To substantiate this viewpoint, studies in the microsporogenesis of these species will be necessary to investigate the normality of the process.

Based on chromosome numbers, Estilai et al. [11], Alberto et al. [15], Ghaffari [17], Foley et al. [19], Al-Turki et al. [22], Nakipoglu [24], Yildiz and Gucel [25], and Marhold and Feliner [26] reported chromosome variations of 2n=12,14,16,18,20,22,24,28,66 and 88 in different Salvia species. This is in agreement with some of the counts in this research.

Al. Turkey et al. [22] and Malalah et al. [23] investigated the basic chromosome number of S. spinosa (x=10). In the present study, S. spinosa revealed 2n=20. Previous reports of S. macrosiphon showed x=10, 2n=20 [9,12] which is in agreement with our results. In this study, the chromosome number of S. atropatana was 2n= 20. The other count of S. atropatana is x= 11 by Afzal-Rafii [8] which is not in agreement with our results. Based on available data, the chromosome count of S. scalrea (2n=22) is in accordance with other results [8,13,27,28,29]. Afzal-Rafii [8], Hedge [30], and Mizianti et al. [31] reported 2n= 12 and 16 for S. nemorosa, but Ghaffari and Sanei Chariat-Panahi [10], Afzal-Rafii [27], Markova and Ivanova [28], Haque [32], Gill [33] and Dobes et al. [34] observed 2n= 14 for this species, which corresponds with our results. Hedge [30] observed 2n=18 for S. virgata. Astonova [35] showed the chromosome count to be 2n= 32, but Afzal-Rafii [8] and Markova and Ivanova [28] displayed 2n= 16 for this species

Table.1. Localities of some Salvia species in Iran.

Species	Locality	Herbarium Code	Height(m)
S. spinosa	Chaharmahal va Bakhtiari- west of Sharekord- south of Dashtak, rousta-e Kaj	HSU (45)	1600
S. spinosa	Chaharmahal va Bakhtiari- west of Shahrekord- Dashtak, north -west of Ghaleh Darvish	HSU (47)	1720
S. spinosa	Chaharmahal va Bakhtiari- north-west of Shahrekord- Chaleshtor	HSU (101)	1900
S. spinosa	Isfahan- south-west of Kashan, Koh-e Dorin	HSU (107)	1800
S. macrosiphon	Isfahan- south of Isfahan, Kolah Ghazi	HSU (63)	1660
S. macrosiphon	Isfahan- south-west of Isfahan, Koh- e Sopheh	HSU (108)	1620
S. macrosiphon	Chaharmahal va Bakhtiari- north-west of Shahrekord- Saman, Ilbagi	HSU (93)	1800
S. atropatana	Chaharmahal va Bakhtiari- south- east of Shahrekord- Borujen- Pir kouh	HSU (36)	2050
S. atropatana	Chaharmahal va Bakhtiari- south- east of Shahrekord, Borujen- Pir kouh	HSU (99)	2550
S. sharifii	Isfahan-south of Isfahan- Kolah Ghazi	HSU (59)	1670
S. sclarea	Chaharmahal va Bakhtiari- south-west of Shahrekord- Boroujen, Vastegan	HSU (7)	1950
S. sclarea	Chaharmahal va Bakhtiari- north-west of Shahrekord, Chaleshtor	HSU (53)	2100
S. nemorosa	Chaharmahal va Bakhtiari- south-west of Shahrekord, toward Izeh	HSU (1)	2000
S. nemorosa	Chaharmahal va Bakhtiari- south od Shahrekord, Ardal- Heydar Abad, Chahar Tagh	HSU (2)	2440
S. nemorosa	Chaharmahal va Bakhtiari- south-east of Shahrekord- Boroujen,Vastegan	HSU (8)	2220
S. nemorosa	Chaharmahal va Bakhtiari- south-east of Shahrekord – Boroujen, Soudejan	HSU (11)	1900
S. virgata	south-west of Shahrekord-Boroujen, Dastgerd	HSU (12)	1800
S. virgata	West of Isfahan-Zayand-e Rod	HSU (78)	2000
S. ceratophylla	Chaharmahal va Bakhtiari- south-east of Shahrekord , Boroujen- Tang-e Sayad, bostan Shir	HSU (28)	2100
S. ceratophylla	Chaharmahal va Bakhtiari- south-east of Shahrekord , Boroujen- Tang-e Sayad, Sephid Dast	HSU (43)	2120
S. ceratophylla	Chaharmahal va Bakhtiari- south-east of Shahrekord , Boroujen- Tang-e Sayad, Sephid Dast	HSU (80)	2080
S. ceratophylla	Chaharmahal va Bakhtiari- south-east of Shahrekord , Boroujen- Tang-e Sayad, Sephid Dast	HSU (81)	2200
S. ceratophylla	Chaharmahal va Bakhtiari- south-east of Shahrekord , Boroujen- Tang-e Sayad, Sephid Dast	HSU (82)	2200

Table2. Chromosome number (2n) Total Haploid Chromosome Length (THCL), Karyotype Formula (KF) and TF% (the Total Form percentage) for Salvia species.

Species	2n	Total Haploid Chromosome Length (µm)	Karyotype Formulae	TF%
S. spinosa	20	0.48-2.25	6st+3t+T	55.9
S. macrosiphon	20	0.4-1.9	M+m+2st+6T	34.03
S. atropatana	20	0.25-0.9	M+sm+2st+2t+4T	45.4
S. sharifii	20	0.35-0.98	2M+2sm+2st+t+3T	44.1
S. sclarea	22	0.3-0.7	2M+m+sm+3st+3t+T	32
S. nemorosa	14	0.38-1.5	sm+2m+t+3T	55.8
S. virgata	16	0.3-1.3	sm+st+t+5T	54.4
S. ceratophylla	14	0.25-1.1	2sm+4t+T	51.6

which is in agreement with the present result. Afzal-Rafii [8,27] studied the chromosome numbers of S. ceratophylla and reported 2n= 44 and 22. In our results, the chromosome number of S. ceratophylla was 2n=14.

Consistent with Ozkan [3], Estilai et al. [11], Ozdemir and Senel [13], Kandemir [14], Ghaffari [17], Ozkan and Soy [18], Tunamoto et al. [36] and Seisuke et al. [37] some of karyological properties in Salvia genus show that some of the species have chromosomes with median point (M), median region (m), submedian region (sm), subterminal region (st) and terminal point (T) centromeres, which support our results. Ozdemir and Senel [13] showed the karyotype of S. sclarea from Turkey which has submedian region centromere in 9th chromosome and the other chromosomes had median region centromere. In this study, the chromosomes of S. sclarea have median point, median region, submedian reigon, subterminal region, terminal region and terminal point centromeres. Therefore, the results obtained in the present paper for this species are not in accordance with those reported by Ozdemir and Senel [13].

The range of chromosome length in Salvia species usually varies between 0.3-3.5 µm [3,11,14,18]. In this study chromosome length varies between 0.25-2.25 µm which is similar to other published results. Also, S. spinosa and S. nemorosa showed the highest symmetrical karyotype (TF%= 55.9 and 55.8, respectively), but S. scalrea showed the lowest symmetrical karyotype (TF%= 32) (Table 2). Indeed, some of the Salvia species have heteromorphic chromosome pairs and each species of Salvia genus generally has different chromosome number [37].

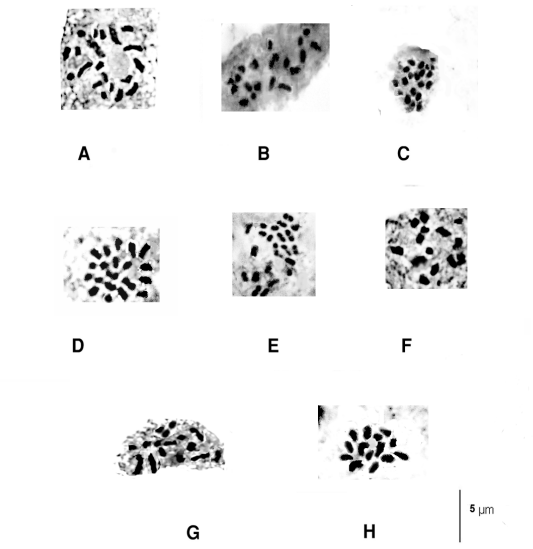

Fig.1. Photomicrograph of mitotic division in somatic cells of Salvia species.
A: S. spinosa (2n=20), B: S. macrosiphon (2n=20), C: S. atropatana (2n=20), D: S. sharifii (2n=20), E: S. sclarea (2n=22), F: S. nemorosa spp. tesquicola (2n=14), G: S. virgata (2n=16), H: S. ceratophylla (2n=14).

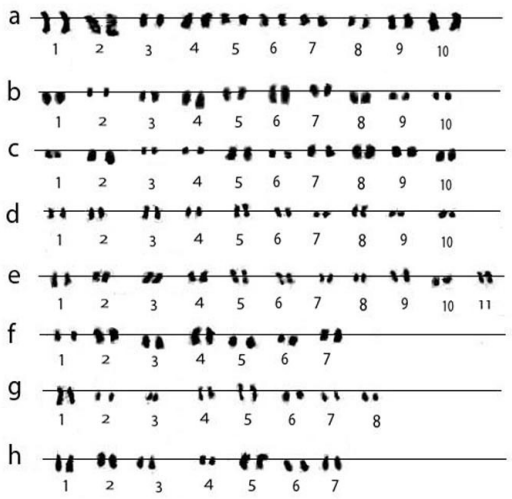

Fig.2. Representative of ideogram in Salvia species.
a: S. spinosa, b: S. macrosiphon, c: S. atropatana, d: S. Sharifii, e: S. sclarea, f: S. nemorosa ssp. tequicola, g: S. virgata, h: S. ceratophylla

CONCLUSION

According to Xun et al. [16] and Al-Turki et al. [22], Salvia is a genus with surprisingly diverse chromosome numbers and in some of the species the variability in chromosome complements is common. Moreover, changes in the chromosome number and variation of karyotype structure can be highlighted as the principal mechanism of species diversification [38]. Identifying the chromosome number of eight Salvia species in this study provides a base for biosystematic studies.

Acknowledgement
Authors are thankful to Dr. C.M. Alberto, University of Argentina for helpful scientific supervision. This study has been supported by the research deputy of Shahrekord University, Shahrekord, Iran with No. 1978.

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