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Research Article

KARYOMORPHOTYPIC STUDIES IN TETRAPLOID VARIETY OF *CYMBOPOGON MARTINII* VAR. *SOFIA*

Thara Saraswathi K.J., Jayalakshmi N.R., *Shivakameshwari M.N.[1] and Jagadishchandra K.S.[2]
Department of Microbiology and Biotechnology, Bangalore University, Bangalore-56
[1]Department of Botany, Bangalore University, Bangalore-56
[2]Department of Botany, Mysore University, Mysore-6
**Author for Correspondence*

ABSTRACT
The present investigation was carried out to study the karyomorphotypic nature of *Cymbopogon martini* var. *Sofia* to determine the ploidy nature. Most of the species of *Cymbopogon* show different ploidy level varying from diploid having 2n=20 chromosomes to hexaploid having 2n=60 chromosomes. The present variety *sofia* collected from Himavadgopala swamy hills is found to be a tetraploid having 2n=40. Based on the chromosome number, ploidy level, chromosome configuration and other meiotic characters x=10 has been considered as the basic number for the genus *Cymbopogon*.

Key Words: *Ideogram, Ploidy, Meiotic Behavior, Cytogenetics, Mitosis*

INTRODUCTION
Cymbopogon martini is a tropical grass found growing wild throughout India and abundantly in South India, particularly in Karnataka state. It serves as a major member with respect to its distribution and economic importance. It is the only species where two remarkably similar forms exist like *sofia* and *motia* (Bor, 1953). Each of these form serve as important source of essential oil which is quite different from other.

C. martini occurs at many ploidy levels varying from diploid (2n=20), tetraploid (2n=40) and hexaploid (2n=60). The tetraploid occurrence is most common, exhibiting polymorphism and supernumerary chromosomes. The first cytological research for the enumeration of chromosome number of Indian *Cymbopogons* was performed by Babu (1936) who observed the diploid number of chromosomes 2n=20 for *C. martinii*. Later, Bor (1953) observed the chromosome number of *C. martini* and also reported as 2n=20 and 40. The variation in ploidy level is seen in majority of wild species distributed in diverse places (Bor 1953).

Raghavan & Arora (1958), Carnahan & Hill (1961) and Mehra *et al.,* (1962) have reported 2n=20 chromosomes for *C. martini* var *Sofia* collected from Coimbatore and Almora. Gupta (1965) has reported the chromosome numbers for eleven species of Indian Cymbopogons collected from various parts of Kerala, out of which five were diploids, three were tetraploids and the rest were hexaploids. Narayan and Jagadishchandra (1966), Gupta (1969a) and Mehra and Sharma (1975) through cytogenetic studies have reported occurance of tetraploid variety from the samples collected from Kemmanagundi (Karantaka), Odakalli (Kerala) and East Himalayan region respectively.

According to Sobti and Lakshmi (1978) the karyotype of *C. martinii* is postulated to be of more evolved type compared to other species in Rusae series. The karyotypic studies made by Jagadishchandra (1976a) on different species of Cymbopogon has revealed the common occurance of metacentric and submetacentric chromosomes with satellite or secondary constrictions confined to only a single pair of chromosome.

The perusal of literature on cytology and cytogenetics in Cymbopogon reveal the chromosome behavior, chiasma frequency with distribution and details on karyomorphological studies, these studies with respect to the Indian species are meager. Thus, a detailed cytologenetical investigation was undertaken in the wild

population of *C. martinii* var. *Sofia* collected near Himavadgopala swamy hills, Chamarajnagar district of Karnataka.

MATERIALS AND METHODS

The wild species of *Cymbopogon martinii* used in the present study were collected from Himavadgopala swamy hills, which are maintained in the germplasm bank at department of Microbiology and Biotechnology, Bangalore University, Bangalore, India. For mitotic study the root tips were collected from plants grown in earthen pots, between 12:30 to 2:30 P.M. The plants were watered two hours before collecting the root tips to make the cells turgid and pretreated with 0.002M of 8-hydroxy quinoline (Tijo and Levan, 1950) for 2½ hours at 15^{O}C.

After thorough washing with running water for 30 mins the roots were fixed in modified Carnoys I fluid (3:1, absolute alcohol: propionic acid) for 24 hours at 15^{O}C and finally the root tips were transferred to 70% ethanol and squash preparations were made. The meiotic preparations were made from spikelets which were harvested between 9:00 to 10:00 A.M. (Sharma & Sharma 1965) and the florets were fixed with modified Carnoys II solution (6:3:1, absolute alcohol: chloroform: propionic acid) for 24 hours at 15^{O}C. The anthers were squashed in a drop of 1% propiono-hematoxilin with a trace of iron alum. The photomicrographs were taken using Carl Zeiss research microscope with apochromatic objective lens system fitted with MF 35mm camera.

RESULTS AND DISCUSSION

Observations

The wild species of *C. martinii* var. *Sofia* revealed tetraploid chromosome number of 2n=40. The somatic chromosomes observed from the root tip cells were generally small. The longest chromosome measured 3.01µm and m-type, the shortest was 1.68µm which was also m-type. Only one pair of submetacentric chromosomes possessed satellites on the short arm. The karyotype formula was $4M+12m+3sm+1sm^{s}$. The karyotype and idiograms are shown in Figure 1. The total chromatin length of the haploid complement was 46.15µm. The karyotype symmetry was 2a type. The details of karyomorphological measurements are given in Table 1.

The meiotic division was regular with n=20 chromosomes showing typical bivalent formation and segregation (Figure 2). Occasionally single tetravalent formation was observed. The chiasmata were localized and distal varied from 1-2 per bivalent.

The mean chaismata per cell was determined to be 35.2 by the observation of 50 PMCs. The frequency of bivalents with single chiasma was 4.8 and that with two chiasmata was 15.2. The second meiotic division was found to be normal with pollen fertility of 83.09%. Chiasma location and frequency were recorded according to Darlington (1965) and a minimum of fifty cells were scored for the analysis. For the comparison of mean chiasma frequency ‘students-t’ test was used.

Discussion

The *C. martinii* var. *Sofia* investigated in the present study displayed a tetraploid chromosome number of 2n=40. Karyomorphologically, the series Rusae is a compact one having only metacentric and submetacentric chromosomes in all the species. The tetraploid variety in the present study possessed four pairs of M-type and 12 pairs of m-type of chromosome with one pair of satellite chromosome of sm type showing ‘2a’ asymmetrical karyotype. These studies are in homology with other tetraploid Cymbopogons investigated earlier in *C. caesius, C. flexuosus, C. polyneuros* (Jagadishchandra 1976a, Sobti & Lakshmi 1978).

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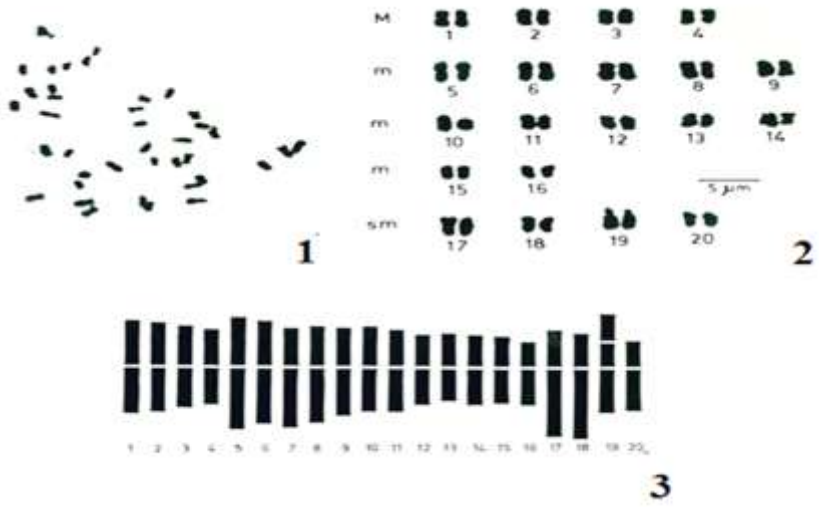
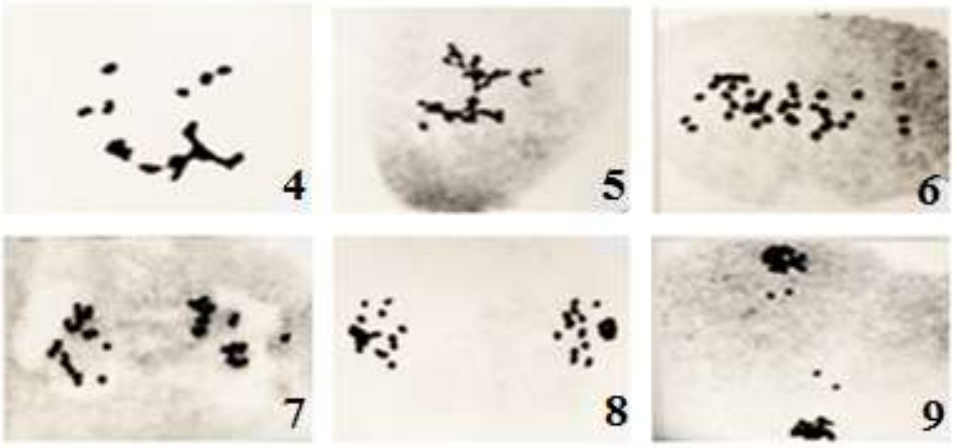
Table 1: Karyomorphological data of *Cymbopogon martinii* variety *Sofia*

Chromosom number	Short arm (µm)	Long arm (µm)	Ratio (la/sa)	TCL	Relative length	Chromosome type
1	1.24	1.24	1.00	2.48	5.37	M
2	1.20	1.20	1.00	2.40	5.20	M
3	1.11	1.11	1.00	2.22	4.81	M
4	1.00	1.00	1.00	2.00	4.33	M
5	1.33	1.68	1.26	3.01	6.52	m
6	1.26	1.57	1.24	2.83	6.13	m
7	1.03	1.64	9.59	2.67	5.79	m
8	1.11	1.48	1.33	2.59	5.61	m
9	1.05	1.28	1.21	2.33	5.04	m
10	1.09	1.22	1.22	2.31	5.00	m
11	0.98	1.20	1.22	2.18	4.72	m
12	0.87	0.98	1.13	1.85	4.00	m
13	0.89	0.92	1.03	1.81	3.92	m
14	0.83	0.95	1.14	1.78	3.86	m
15	0.81	0.97	1.19	1.78	3.86	m
16	0.67	1.01	1.50	1.68	3.64	m
17	1.00	1.89	1.89	2.89	6.26	sm
18	0.89	1.94	2.17	2.83	6.13	sm
19	0.66	1.19	1.80	2.55	5.53	sm
20	0.70	1.26	1.80	1.96	4.25	sm

Absolute chromatin length: 46.15µm; Karyotype formula: 4M+12m+3sm+1sm^{s}; Karyotype asymmetry: 2a



Figures 1-3: Karyomorphology of *C. martinii* 1. Mitotic metaphase plate. 2. Karyotype. 3. Idiogram



Figures 4-9: Meiosis in *C. martinii*. 4. Metaphase-I, clumping. 5. Early anaphase-I, stickiness. 6, 7, 8. Anaphase-I, showing various degrees of stickiness. 9. Late anaphase-I, showing 20-20 segregation

In the presently investigated variety, the chiasmata were observed to be localized and distal. The chaismata in plants are random or localized. The chaismata confined to short region near noncentric end of the chromosome are recognized as distal ones (John & Lewis 1965). The cause of localization in plants however varies. Darlington (1935) proposed that the region pairing first is preferred for chaisma formation and the heterochromatic segments influencing pairing mechanism also influences chiasma formation. Similar chaisma formation pattern might have been occurred in the presently investigated variety. During the present study lower mean chaismata per bivalent was observed and higher mean chaismata per bivalent occur only in diploid species of Cymbopogon (Snyder, 1961). Present investigations on meiotic behavior of *C. martini* revealed that, meiosis I and II were normal with no distinct chromosomal irregularities, in accordance with the earlier studies (Gupta, 1970d & 1970e).

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REFERENCES
Babu CN (1936). Chromosome numbers in Cymbopogon species. *Current Science* **4** 739-740 and **4** 874-875.
Bor NL (1953). The genus Cymbopogon Spreng. In India Burma and Ceylon Part I. *Journal of the Bombay Natural History Society* **51** 890-916.
Carnahan HL and Hill HD (1961). Cytology and genetics of forage grasses. *Botanical Review* **27** 1-162.
Darlington CD (1935). The time place and action of crossing over. *Journal of Genetics* **31** 185-212.
Darlington CD (1965). Recent advances in Cytology. *J &A Churchill Ltd* London 85-134.
Gupta BK (1965). Chromosome numbers in some species of Cymbopogon. *Proceedings of the National Academy of Sciences, India Section B* **62** 155-157.
Gupta BK (1969a). Studies in the genus Cymbopogon Spreng I Chromosome studies in Indian Cymbopogons. *Proceedings of the National Academy of Sciences, India Section B* **70** 241-247.
Gupta BK (1970d). Studies in the genus Cymbopogon Spreng VI A new name in genus Cymbopogon. *Proceedings of the National Academy of Sciences, India Section B* **71** 90-93.
Gupta BK (1970e). Studies in the genus Cymbopogon Spreng VII Some varieties in Indian Cymbopogons. *Proceedings of the National Academy of Sciences, India Section B* **71** 94-99.

Jagadishchandra KS (1976a). Reproductive behavior of some species of Cymbopogon (Poaceae). In: *Embryology of crop plants Indo-soviet symposium,* New Delhi 12-13.
John B and Lewis KR (1965). The meiotic system *Protoplasmatologia* Springer-Verlag New York.
Mehra KL, Subramanyam KN and Swaminathan MS (1962). Cytological studies in some members of Andropogoneae. *Journal of the Indian Botanical Society* **41** 491-503.
Mehra PN and Sharma ML (1975). Cytological studies in some Central and Eastern Himalayan grassed I The Aniropogoneae. *Cytologia* **40** 61-74.
Narayan KN and Jagadishchandra KS (1966). Cytological studies in species of Cymbopogon from South India. *Journal of Mysore University* **20** 66-72.
Raghavan R and Arora MC (1958). Chromosome numbers in Indian medicinal plants-II. *Proceedings of the National Academy of Sciences, India Section B* **47** 352-358.
Sharma and Sharma (1965). Chromosome techniques Theory and practices. *Butterworths* London.
Snyder LA (1961). Asyndesis and meiotic non-reduction in microsporogenesis of apomictic *Paspalum secans*. *Cytologia* **26** 50-61.
Sobti SN and Lakshmi B (1978). Karyomorphological studies in the genus Cymbopogon. In: *All India high level symposium on researches in plant cytogenetics.*
Tijo JH and Levan A (1950). The use of oxyquinoline in chromosome analysis. *Ann Estae Exot Aula Dei* **2** 21-64.