

Karyomorphological study of *Blumea aromatica* DC: An ethnobotanical species of Manipuri community of Tripura, India

H. Reshmi Singha, Sunita Debbarma, Jayasri Sinha, Sangram Sinha and Rabindra Kumar Sinha*

Cytogenetics and Plant Biotechnology Laboratory, Department of Botany, Tripura University (A Central University), Suryamaninagar, Tripura

ABSTRACT

Blumea aromatica DC is an semi-wild species recorded in Tripura under the family Asteraceae. Morphologically the species is characterized by having winged petiole or leaf appendages along with membranous leaf. Detailed karyomorphology of the species is worked out and the karyotype formula is constructed as $A_2B_4C_{10}D_2$ with somatic chromosome number $2n = 18$. Regional significance of *Blumea aromatica* is also recorded and documented as an ethno-botanical species in the present investigation.

Key words: *Blumea aromatica* DC; Karyomorphology; ethno-botanical species

INTRODUCTION

Tripura is one of the smallest State of North East India and inhabited by the Tribal of Tibeto-Burman stock with as many as 19 different tribal communities [1]. Tripura is also represented by other communities like Manipuris and Bengali since king dynasty in the State. This ethological diversity has not only reflected their sociological and cultural aspects but also revealed unique traditional food habit and rural-herbal therapy, [2]. The state is also rich in floristic diversity [3]. Many of these ethno-botanical species and their traditional knowledge of uses associated with health care system are to be explored scientifically. *Blumea aromatica* DC under the family Asteraceae is reported to be ethno-botanical significance in this region [2]. The species is very familiar to Manipuri community and called as 'Leikhaman'. Fresh and fully expanded leaves are used as vegetables and leafy spice. In spite of the significant traditional value no attempt has been taken to carry out cytological analysis of the species growing wild and semi-wild condition in this north-eastern part of Tripura. In view of the above context an effort has been made to study karyomorphological nature of the species at cytological level along with the morphometrical characters of the plant habit.

MATERIALS AND METHODS

Plant specimen (*B. aromatica*) was collected from the rural and Manipuri community based areas of Tripura. Plant was accordingly described and recorded. Finally the plant was identified consulting authentic floristic literatures like Flora of Tripura [3], Flora of Assam [4] and Flora of British-India [5]. Cross examination of the species was done consulting with Prof. B.K. Datta, Plant Taxonomy and Biodiversity Laboratory, Department of Botany, Tripura University.

Shoot tips of *Blumea aromatica* were collected from healthy plants for cytological studies. Outer covering of young leaves of shoot tip were removed and extreme tips measuring 0.5-1.0 mm were pre-treated with saturated pDB (para-dichlorobenzene) solution in freezing temperature (4°C) for 5 minutes and finally in 12°C for 3.30 hours. Then the shoot tips were fixed in solution of acetic acid and ethyl alcohol mixture (1:3) at room temperature for overnight. After that they were treated with 45% acetic acid for 10 minutes and stained in a solution of 2% aceto-orcein and (N) HCl (9:1) mixture with warming for 3-4 seconds. Staining was carried out for overnight. After

staining the shoot tips were squashed in 45% acetic acid and studied under compound microscope. At least 25 observations were made from such treatments. Chromosomes of the plates were drawn using oil immersion objective (x 1360). Measurement of chromosomal complements like absolute length of the chromosome, length of short arm and long arm and F% are carried out for each complement with 5 different metaphase plates. Detailed karyotype table was constructed from the well spread metaphase plates using total length, short arm, long arm and F% of the chromosome complements and types of the chromosome complements. The F% was determined by using the following formula:

$$F\% = \frac{\text{Short arm length}}{\text{Total length of chromosome}} \times 100$$

Total Forma percentage (TF %) of chromosomal complements [6] was also calculated.

RESULTS AND DISCUSSION

Plants are perennial shrubs and attaining the height upto ~ 130cm or more. The stem is round, green, glandular, pubescent and aromatic. Leaves are simple, alternate, membranous, scabrous, lanceolate and toothed. Mature leaves measuring 21-26 x 4.9-7 cm in size and petiolated while young leaves are crowded at the top and sessile (Fig. 1). Lateral nerves are 14 -18 on either half of the leaf. The leaf is also characterized by the presence of leaf appendages at the leaf base (Fig. 2). Size of the leaf appendages varied from 0.2-0.8 cm in length. Presence of leaf appendages in the mature leaves along with sessile petiole in younger ones is a characteristic feature of *B. aromatica* [5]. Flowers are in capitulum and ~0.9 cm long, inflorescence bearing flowers are in a long stalk measuring 53.2 ± 14.84 cm. The length of inflorescence and inflorescence stalk are found to be 0.9 ± 0.02 cm and 0.31 ± 0.01 cm respectively. Flowers are characterized by the ray and disc florets, a characteristic feature of the Asteraceae. Ray florets are the outer female flowers and ranging from 170 - 180 in numbers, corolla is filiform and minutely 2 – 3 lobed, yellowish white ~ 0.5 cm long; disc florets are the inner bisexual flowers and ranging from 13 – 18 in numbers, regular, epigynous, corolla tubular, slender, 5 lobed, yellowish white, ~ 0.5 cm long. The corolla lobes are glandular. Seven types of bracts are observed in each inflorescence. The morphometric characters of the flower is worked out and presented in table 1 and 2. In each flower there are 5 stamens, anthers saggitate, 2 mm long, filaments are ~ 4 mm long, pollens spiny (0.02 x 0.02) mm in size. Carpels are 2, syncarpous, ovary inferior, style long, stigma bifurcated. The fruits are cypsela, small, densely pilose and 10 ribbed. The minute fruit size is found to be 0.24 x 0.06 mm (length x breadth) with distinct white pappus of 18.3 ± 0.48 numbers per fruit. The pappus is 4.86 x 0.02 mm (length x breadth) in size.

From the well spread metaphase plates somatic chromosome number of the species is determined and found to be $2n = 18$ (fig.3). This finding is also collaborated with previous observation [7, 8]. However, range in variation of somatic chromosome number $2n=16$ in *B. barbata* to $2n=36$ in *B. lacera* with B-chromosomes has been reported [9]. In the present investigation all the metaphases revealed homogenously 18 somatic chromosome number. Characteristics measurements of chromosomal complements are presented in tabular form (table,3). From the 5 different chromosomal measurements average value of each complement was determined, and presented in table-3. Absolute size of the chromosomes ranged from $2.646 \pm 0.36 \mu\text{m}$ to $4.262 \pm 0.854 \mu\text{m}$ with a pair of chromosomes having secondary constriction (table, 3). The secondary constriction is associated with the short arm of the chromosomal complement having an absolute length of $3.89 \pm 0.37 \mu\text{m}$. The total length of haploid chromosomal complement is $30.65 \mu\text{m}$ and the total forma percentage (TF %) is 42.43. On the basis of primary constriction and size of chromosomal complements, the chromosomes are classified into four morphological types:

Type A: Chromosomes with an average size of $3.896 \pm 0.373 \mu\text{m}$ having nearly median(nm) primary constriction and sub-terminal(st) secondary constriction.

Type B: Chromosomes with an average size range of $3.088 \pm 0.296 \mu\text{m}$ to $3.528 \pm 0.72 \mu\text{m}$ having typical metacentric (m) in nature.

Type C: Chromosomes with size range of $2.646 \pm 0.36 \mu\text{m}$ to $4.262 \pm 0.854 \mu\text{m}$ and bears nearly median (nm) primary constriction.

Type D: Chromosomes with an average size of $2.646 \pm 0.36 \mu\text{m}$ having submedian (sm) primary constriction.

Based on chromosomal typification, the karyotype formula of *B. aromatica* is found to be $A_2 B_4 C_{10} D_2$. The respective karyogram of the karyotype formula is presented in Fig.5 The somatic chromosome of *B. aromatica* show

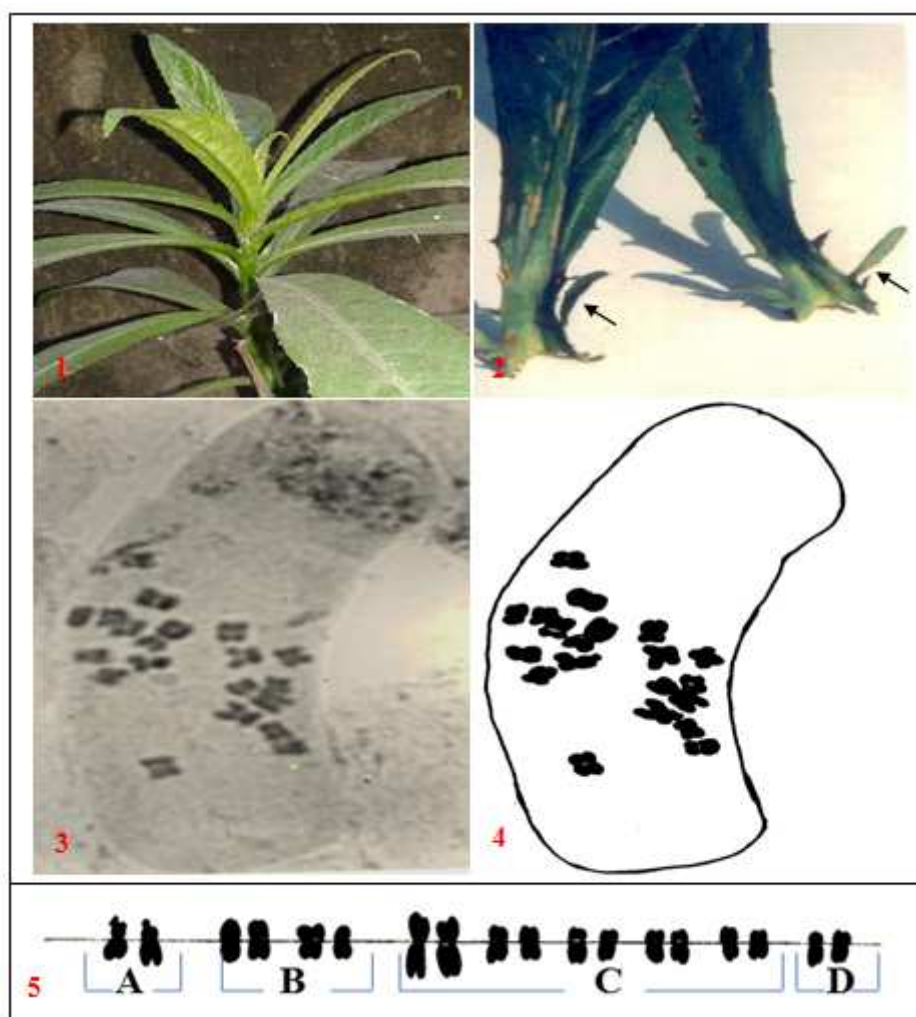
homogeneity with slight size variation. The TF% value indicates symmetry of chromosomes to a certain extent. According to degree of asymmetry, the karyotype of the species (table,4) belongs to the category 3A [10]. Relatively less size differences between the chromosomes of the complement suggests its karyotype is to be considered as slightly asymmetric one. The chromosome count and constancy of the karyotype recorded in the present taxon could be utilised to measure the cytological as well as karyomorphological variability among the ecotypic population of the species.

Table 1. Detail morphometric characters of the individual flower

Categories of bracts (length x breadth) mm	No. of bracts /inflorescence (mean \pm SD)	No. of ray florets (mean \pm SD)	Length of ray florets (mean \pm SD) mm	No. of disc florets (mean \pm SD)	Length of disc florets (mean \pm SD) mm
7.00 x 0.12	19.00 \pm 2.90	172.46 \pm 3.80	6.8 \pm 0.23	15.26 \pm 1.43	6.8 \pm 0.23
5.90 x 0.16	12.26 \pm 0.45				
5.00 x 0.08	08.13 \pm 0.56				
4.00 x 0.08	08.20 \pm 0.90				
3.00 x 0.08	09.20 \pm 1.74				
2.13 x 0.05	08.73 \pm 1.49				
1.13 x 0.04	07.90 \pm 0.79				

Table 2. Morphometric measurement of reproductive parts

Length of corolla tube (mean \pm SD) mm	Length of style (Mean \pm SD) mm	Length of stigma (mean \pm SD) mm	Length of filament (mean \pm SD) mm	Length of anther (mean \pm SD) mm	Length of ovary (mean \pm SD) mm
4.86 \pm 0.22	5.36 \pm 0.22	0.5 \pm 0.00	3.86 \pm 0.00	2 \pm 0.00	1 \pm 0.00



Figures: 1) Part of mature vegetative Shoot of *B. aromatica* DC, 2) Petiolar base of *B. aromatica* DC leaves showing characteristic leaf appendages (), 3) Somatic metaphase plate $2n=18$, 4) Camera Lucida drawing of the metaphase plate (Magnification X1360); 5) Karyogram of *B. aromatic* DC showing chromosome types

Table 3. Karyomorphological characteristics of the *Blumea aromatica* DC.

Chromo- some pair	Karyomorphological parameters of <i>B. aromatica</i>						
	Absolute length ($\mu\text{m} \pm \text{SD}$)	Long arm ($\mu\text{m} \pm \text{SD}$)	Short arm ($\mu\text{m} \pm \text{SD}$)	Arm ratio	F%	Nature of chromo-some	Type
1	3.90 \pm 0.37	1.91 \pm 0.28 3.38 \pm 0.28	0.52 \pm 0.18 1.47 \pm 0.00	7.26 \pm 2.15	38.08 \pm 3.79 13.05 \pm 3.68	nm	A
2	3.09 \pm 0.30	1.54 \pm 0.15	1.54 \pm 0.15	1.00 \pm 0.00	50.00 \pm 0.00	m	B
3	3.53 \pm 0.72	1.76 \pm 0.36	1.76 \pm 0.36	1.00 \pm 0.00	50.00 \pm 0.00	m	B
4	4.26 \pm 0.85	2.43 \pm 0.55	1.84 \pm 0.33	1.31 \pm 0.12	42.67 \pm 1.68	nm	C
5	3.68 \pm 0.00	2.21 \pm 0.00	1.47 \pm 0.00	1.50 \pm 0.00	39.94 \pm 0.00	nm	C
6	3.90 \pm 0.18	2.43 \pm 0.18	1.47 \pm 0.00	1.64 \pm 0.12	37.80 \pm 1.74	nm	C
7	3.01 \pm 0.36	1.69 \pm 0.18	1.32 \pm 0.181	1.28 \pm 0.04	43.76 \pm 0.79	nm	C
8	2.65 \pm 0.36	1.52 \pm 0.16	1.12 \pm 0.20	1.37 \pm 0.13	42.16 \pm 2.17	nm	C
9	2.65 \pm 0.36	1.65 \pm 0.23	0.99 \pm 0.13	1.66 \pm 0.02	37.51 \pm 0.13	sm	D

Table 4. Stebbins catagorization of karyotype *Blumea aromatica* DC

Ratio Largest/Smallest	Proportion of the chromosome with arm ratio <2:1			
	0.00	0.01-0.50	0.51-0.90	1.00
<2:1	1A	2A	3A <i>B. aromatica</i> 3A	4A

CONCLUSION

Blumea aromatica is a common ethnobotanical homestead species to Manipuri community of Tripura. First karyomorphological account of the taxon with a karyotype formula $A_2B_4C_{10}D_2$ are described in the present communication.

Acknowledgement

Authors are thankful to the University Grants Commission, (UGC) New Delhi for providing financial support to the Department of Botany, Tripura University, Suryamaninagar-799022, India.

REFERENCES

- [1] Deb Varman SBK, *The Tribals of Tripura a dissertation*. Spl Series 1, Directorate of Research Publ. Governmentt of Tripura, Agartala, India, **1986**, 113.
- [2] Sinha RK, Chakraborty S, K. Roy, Sinha S, In: Inter.conf. on promotion and development of Botanicals, P.K. Mukharjee (Eds.), **2005**, 272-275.
- [3] Deb DB, *The Flora of Tripura*. Today and tomorrow's Publ., New Delhi, **1983**
- [4] Kangilal PC, Dev RN, *Flora of Assam*. Omsons Publ., New Delhi, **1939**, 113.
- [5] Hooker JD, *The Flora of British India*, SIC, L. Reeve & Co., London, **1882**.3, 260-270.
- [6] Levan AK, Fredgra K and Sandberg A, *Hereditas*, **1964**, 52, 201-220.
- [7] Malla SB, Bhattarai S, Gorkhali M, Saiju H, Kayastha M, Singh MP, *Taxon*, **1977**, 26: 557-565.
- [8] Peng IC, Hsu CC, *Taxon*, **1978**, 26: 557-565 .
- [9] Methew A and Methew PM, *Cytologia*, **1975**, 40, 365-370.
- [10] GL Stebbins, *Chromosomal evolution in Higher plant*, 1971, Edward Arnold., London