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KARYOTYPIC DIVERSITY AMONG EIGHT *ZEA MAYS* L. VARIETIES

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Abstract

Staining property of interphase nuclei and prophase chromosomes, diploid chromosome number, total chromosome length (TCL), symmetric and asymmetric indices of karyotype were studied in eight maize varieties released by BARI. 2n = 20 chromosomes were found in Barnali, Mohor, Khoi Vhutta, BS-1, B-5 and BM-7 whereas 2n = 22 chromosomes in China and 2n = 24 chromosomes in B-73 were observed. TCL was highest in Mohor (190.49 ± 5.61 µm) and lowest in B-73 (69.30 ± 2.51 µm). These varieties showed significant variation in cytogenetical parameters. Results obtained are expected to supplement genetic identification of maize varieties in variety conservation efforts.

Maize (*Zea mays* L.) belongs to Poaceae, originated from the lowland tropics in South America and is now cultivated nearly all over the world (Pokhmelnykh and Shumny 2003). Bangladesh Agricultural Research Institute (BARI) has released a number of varieties such as Barnali, Shuvra, Khoi Vhutta, Mohor, BARI vhutta-5, BARI vhutta-6, BARI vhutta-7, BS-1, B-5, BM-7, B-73 and 11 hybrids of this species (BARI hybrid 1-11) (Mondal *et al.* 2014). These varieties with many lines are characterized on the basis of their morphological features. This kind of characterization sometimes creates problem since phenotypic features are not always reliable. For this purpose, an authentic characterization and streamlining of these germplasm are needed to avoid overlapping and misidentification during varieties conservation. A number of previous reports are available on this species concerning somatic chromosome number with various banding procedure (Kuwada 1915, Raicu and Chirila 1971, Newell and Wet 1973, Stalker *et al.* 1977, Petrov *et al.* 1979, Sarma and Sharma 1986, Sokolov *et al.* 1998, Molina and García 1999, Pokhmelnykh and Shumny 2003). Moreover, no cytogenetical work of this species was conducted in Bangladesh previously. In the present study, a conventional karyotype analysis was made to characterize and evaluate the karyotypic diversity among eight maize varieties, namely Barnali, Mohor, Khoi Vhutta, China, BS-1, B-5, BM-7 and B-73 released from BARI.

Root tips were collected from plants growing in garden soil and pre-treated with 8-hydroxyquinoline (0.002%) for 2 hrs 30 min at 18°C followed by 15 min fixation in 45% acetic acid at 4°C and preserved in 70% alcohol for future use. Then Orcein-stained chromosomes were observed under the compound microscope. Karyotypic formula was determined on the basis of number of chromosome and position of centromere (Levan *et al.* 1964). Various karyomorphological parameters and asymmetry indices calculated with the help of KaryoType software (Altinordu *et al.* 2016). Results are shown in Table 1.

On the basis of staining property of interphase nuclei and prophase chromosomes three groups could be recognized: Firstly, heterochromatins firmly aggregated in the interphase nuclei and then was homogenously distributed in the prophase chromosomes; secondly, heterochromatins distributed homogenously in interphase nuclei and in the prophase chromosomes and thirdly, aggregated heterochromatins present in both interphase nuclei and prophase chromosomes.

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Table 1. Comparative karyosystematic analysis of eight *Zea mays* varieties.

Features	Barnali	Mohor	Khoi	China	BS-1	B-5	BM-7	B-73
2n	20	20	20	22	20	20	20	24
Satellite	-	-	3	1	-	-	2	-
CF	16 m + 4 sm	20 m	18 m + 2 sm	16 m + 6 sm	20 m	16 m + 4 sm	16 m + 4 sm	24 m
TCL (µm)	183.89 ± 4.03	190.49 ± 5.61	181.77 ± 2.87	147.48 ± 3.89	133.06 ± 4.71	137.75 ± 5.19	149.62 ± 5.43	69.30 ± 2.51
ACL (µm)	9.19	9.52	9.09	6.70	6.65	6.89	7.48	2.89
CV_{CI}	11.65	6.13	9.42	14.39	7.80	13.24	10.83	3.01
CV_{CL}	23.60	26.64	13.98	23.24	14.15	23.46	14.51	11.80
M_{CA}	13.52	9.14	14.11	13.92	13.90	16.14	21.71	9.68
AsK %	56.33	54.91	57.28	57.60	56.93	58.34	61.29	54.85
TF %	43.67	45.09	42.72	42.40	43.07	41.66	38.71	45.15
Syi %	77.51	82.12	74.59	73.61	75.64	71.42	63.16	82.32
Rec %	71.38	56.59	79.96	72.49	81.73	70.44	79.33	82.05
A_1	0.23	0.16	0.24	0.23	0.24	0.26	0.35	0.18
A_2	0.24	0.27	0.14	0.23	0.14	0.23	0.15	0.12
A	0.14	0.09	0.14	0.14	0.14	0.16	0.22	0.10
AI	2.75	1.63	1.32	3.34	1.10	3.11	1.57	0.35
DI	9.82	11.88	5.70	10.21	5.98	8.99	5.64	5.46
Category	1B	1B	1A	2B	1A	2B	2A	1A

2n = Somatic chromosome number, CF = Centromeric formula, TCL = Total chromosome length (µm), ACL = Average chromosome length (µm), CV_{CI} = Coefficient of variation of centromeric index, CV_{CL} = Coefficient of variation of chromosome length, M_{CA} = Mean centromeric asymmetry, AsK % = Karyotype asymmetry index (%), TF % = Total form value (%), Syi % = Karyotype symmetry index (%), Rec % = The index of chromosomal size resemblance, A_1 = Intrachromosomal asymmetry index, A_2 = Interchromosomal asymmetry index, A = Degree of asymmetry of karyotypes, AI = The asymmetry index, DI = The dispersion index, m = Metacentric chromosome, sm = Sub-metacentric chromosome.

Somatic chromosome numbers were 20, 22 and 24 in different varieties (Fig. 1). However, variation in somatic number (2n = 21, 22, 24, 27, 38, 39, 40, 42, 46, 48, 52, 56, 58 and 82) in maize have been reported by other authors (Kuwada 1915, Raicu and Chirila 1971, Newell and Wet 1973, Stalker *et al.* 1977, Petrov *et al.* 1979, Sarma and Sharma 1986, Sokolov *et al.* 1998, Molina and García 1999, Pokhmelnykh and Shumny 2003). The above data indicate that polyploidy, aneuploidy and intraspecific hybridization play a key role in the origin of series of different chromosome numbers for this species. Karyotypes of these eight varieties had predominantly metacentric (m) chromosomes (Table 1). Data presented in Table 1 also show great variability in TCL and ACL. Satellites were detected in three varieties of which Khoi Vhutta had three satellites (two in pair V and one in pair III); variety China showed one in chromosome pair V and BM-7 had a pair of satellite in pair IX. All the observed satellites were positioned at the short arms of the respective chromosomes (Fig. 1). According to the Stebbins's (1971) classification, Khoi Vhutta, BS-1 and B-73 were positioned in 1A whereas BM-7 was placed in 2A karyotype. Barnali and Mohor showed 1B karyotype. In contrast, 2B karyotype was found in China and B-5 (Table 1).

Fig. 1. Karyotypes prepared form Orcein-stained mitotic metaphase chromosomes of eight *Zea mays*. varieties. (a) Barnali, (b) Mohor, (c) Khoi Vhutta, (d) China, (e) BS-1, (f) B-5, (g) BM-7 and (h) B-73. Bar = 10 µm.

Karyosystematics is pertinent feature in evaluation of genetic relationship and divergence among species or populations (Guerra 2008). The AI of analyzed varieties had very variable values, ranging from 0.35 - 3.34 and the CV_{CI} of these eight varieties ranging from 3.01 - 14.39. Higher AI and CV_{CI} value represent asymmetric karyotype whereas lower values indicate symmetric nature. From the data considering AI and CV_{CI} values, it can be said that five varieties

(B-73, BS-1, Khoi Vhutta, BM-7 and Mohor) were of symmetric karyotypes and their AI values ranged from 0.35 - 1.63 and CV_{CI} values ranged from 3.01 - 10.83. The rest three varieties with asymmetric karyotypes had AI values between 2.75 and 3.34 and CV_{CI} values between 11.65 and 14.39. According to these values, while the most asymmetric karyotype was seen in China and the most symmetric one belonged to B-73. According to AI *versus* CV_{CI} analysis, all the varieties showed a systemic appearance and heterogeneity with respect to their asymmetry indices. Varieties with symmetrical karyotypes were placed in one group while those with asymmetrical karyotypes were positioned in another. Thus, varieties of the first group were primitive and those in second group are advanced (Stebbins 1971). Considering the A_1 and A_2 values, all the species displayed a moderate variation level ranging from 0.16 - 0.35 in case of A_1 and 0.12 - 0.27 for A_2 (Table 1). Dispersion index (DI) was also calculated to determine the karyotype asymmetry of the analyzed varieties and it was found that DI values were higher in the varieties with asymmetric karyotype and lower in the varieties with symmetric karyotype (except Mohor). The B-73 variety is characterized by the lowest TCL, CV_{CI}, CV_{CL}, AsK%, AI and DI values than the rest of varieties. Present findings suggested that B-73 were distantly related having different somatic chromosome number (2n = 24) and smaller chromosomes (ACL 2.89 µm) compared to the rest studied varieties (ACL ranging from 6.65 - 9.52 µm) of *Z. mays*. The karyotypes of other seven varieties except B-73 displayed relatively close relationship in respect of different karyotypic parameters. According to those studied chromosomal indices, the highest similarities of chromosomal data showed in between China and B-5. In addition, Barnali and Khoi vhutta showed proximity in chromosomal characteristics (Table 1).

The investigation has elucidated the chromosomal features of eight varieties of *Z. mays*. The presence of both symmetrical and asymmetrical karyotypes indicates the chromosomal rearrangements that occurred in different evolutionary steps in this species. The karyomorphological study of these eight *Zea mays* varieties released by Bangladesh Agricultural Research Institute may provide helpful information in further breeding program in Bangladesh.

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