IMPACT: International Journal of Research in Applied, Natural and Social Sciences (IMPACT: IJRANSS)
ISSN(P): 2347-4580;ISSN(E): 2321-8851
Vol. 4, Issue 7, Jul 2016, 127-132

KARYOTYPE ANALYSIS IN *ZEA MAYS* L. VAR. EVERTA (POPCORN) CULTIVATED WITHIN OWERRI, SOUTHEAST NIGERIA

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ABSTRACT

Karyotype analysis of *Zea mays* L. variety everta (Popcorn) was carried out with an aim of establishing any karyotypic features of this plant which could be useful in designing crop improvement efforts for the crop. Fruits of the plant were obtained from operators and certified in a University Herbarium. Germinating root tips were pretreated in colchicine solution and fixed with acetic ethanol. Metaphase chromosomes were accessed after hydrolysis and squashing in orcein. A diploid chromosome count of 2N = 20 was recorded for all cells. The karyotype was highly asymmetrical with intrachromosomal asymmetry index of 0.4548 and interchromosomal asymmetry index of 0.4314. The shortarm had a very high coefficient of variation value of 84.14%, while total length coefficient of variation value was 43.14%. The implication of a history of both major and cryptic activities in this karyotype can be interpreted as karyotypic instability which could be exploited in improvement regimes such as recurrent selection.

KEYWORDS: Everta, Karyotype, Heterogenous, Asymmetrical, Selection

INTRODUCTION

The maize plant (*Zea mays* L.) ranks as a top staple and cereal crop in Sub-Saharan Africa, serving as food for both Humans and animals and producing raw material for industries. Maize has been estimated to contain about 70.7 percent Carbohydrate, 10 percent Protein, 4 percent Oil, 13.5 percent moisture, 1.4 percent ash and 0.4 percent of other substances Saini and Mathauda (2013). Maize and maize products constitute raw materials for many Agro-based industries leading to production of starch, dextrose, corn syrup, corn oil, cornflakes, popcorn etc.

The use of *Z. mays* var. everta, popularly called popcorn has witnessed increased popularity in recent times in Nigeria. In many city centers, recreation facilities and school premises, 'popcorn joints' are springing up more often. The popularity of this variety, especially among children of school age, stems from the fact that the kernel expands and pops out when subjected to heat. The heat induces pressure in the hard sealed hull and the densely packed starchy interior explodes consequently, making a mild popping sound, from where it derives its name.

MATERIALS AND METHODS

Some grains of *zea mays* var. everta was obtained from popcorn operators within Owerri, Nigeria. The fruits were certified at the herbarium of the Department of Plant science and Biotechnology, Imo State University Owerri. Fruits were then germinated in petri-dishes using wet cotton wool. Germinating root tips were pretreated in 0.05 percent colchicine solution for 5 hours before fixation in acetic ethanol (1:3 v/v) for 24 hours. They were then stored in 70 percent ethanol in

the refrigerator at 4^{0C}. During cytological investigation, stored root tips were hydrolysed in 1N HCL in a water bath for 8 - 10 minutes, stained and squashed in 2 drops of two percent orcein, mounted and viewed. Metaphase chromosome count was carried out and good spreads were photomicrographed.

Chromosome length measurement was carried out with appropriate eyepiece graticles. Data such as long arm, short arm, total length, centromere position, presence or otherwise of satellites and other accessories were collected. Other data such as centromeric index, arm ratio, total form percent and asymmetry indices were worked out.

RESULTS

In all metaphase stage cells viewed, the diploid chromosome number was 2N = 20 (Plate 1). The chromosomes were regularly distributed within the plate and were moderately sized with no supernumeraries observed. The longest chromosome measured 5.87µm and was 18.75 percent of the total chromatin length (TCL). The shortest chromosome was 1.92 µm and 6.14 percent of the total length. The chromosome formula was 2n = 2x = 20 = 0M + 3m + 6sm + 1^{st}; with a mean centromeric index of 0.3339. The values for the coefficient of variations for short-arm, long-arm and total length were 84.15 percent, 25.92 percent and 43.14 percent respectively. An outlay of the karyotype data for each of the 10 homologues in the karyotype is presented in Table 1, while Table 2 gives a summary of the karyotype parameter indices for the plant.

Plate 1: Karyotype of Zea mays var. everta (2N = 20)

Table1: Karyotype Data of Zea Mays Var. Everta Showing Homologue Dimensions

H.	L/ARM	S/ARM	TOTAL	D.	A.RATIO	C.I	C. LOC.	2N
1	2.94	2.93	5.87	0.01	1	0.4991	M	20
11	2.63	2.63	5.26	0.00	1.0	0.5000	M	‘’
111	2.46	0,83	3.29	1.63	2.96	0.2523	SM	‘’
IV	2.19	0.72	3.29	2.91	1.47	0.2466	SM	‘’
V	1.39	1.37	2.76	0.02	1.01	0,4964	M	‘’
VI	1.89	0.63	2.52	1.26	3.00	0.2500	SM	‘’
VII	1.80	0.60	2.40	1.20	3.00	0.2500	SM	‘’
VIII	1.98	0.30	2.28	1.68	6.6	0.1515	ST	‘’
IX	1.53	0.55	2.08	0.98	2.78	0.3595	SM	‘’
X	1.44	0.48	1.92	0.96	3.00	0.3333	SM	‘’
TOTAL	**20.25**	**11.04**	**31.29**	**9.21**	**27.39**	**3.3387**		

Key: H = Homologue, L = Long, S = Short, D = Difference, A = Arm, C.I = Centromeric Index

Table 2: Summary of Parameter Indices of *Zea mays* var. everta

Parameter	Value
∑L	20.25 µm
∑S	11.04µm
TCL	31.29 ‘’
Č	3.13µm
S	1.35
S^2	1.83
TF%	36.11
A_1	0.4548
A_2	0.4314
CV (Shortarm)	84.14%
CV (Longarm)	25.92%
CV (Total)	43.14%

Key: ∑L – Longarm summation, ∑S – Shortarm summation, TCL – Total chromatin length, Č – Mean length, S – Standard deviation, S^2 – Variance, TF% - Total form percent, A_1 – Intra chromosomal asymmetry index, A_2 – Interchromosomal asymmetry index, CV – Coefficient of variation

DISCUSSIONS

The karyotype of an organism is defined to include but not limited to the following components; Chromosome number, chromosome length, location of the centromere, presence or otherwise of satellites within the complement. Chromosome number constitutes the most significant cytological character of the species, hence its role in cytotaxonomy and related fields. In the present investigation the diploid chromosome count was 2n = 20. This result agrees with reported chromosome numbers in the species *Zea mays* (Rhoades and McClintock, 1935; McClintock, 1948; Ting, 1958; Wolfe *et al*.,1989; Gaut and Doebley, 1997; Mehra and Sharma, 1975; Kato, *et al*., 2004; Albert, *et al*., 2010; Sadder and Weber, 2001). This result reinforces the close morphological similarities existing among the varieties in the species. But it fails to explain the phenotypic dissimilarity as regards seed texture which distinguishes the variety everta from other varieties of the species. The observation as reported by Kuwada (1919) that variations were evident in the number of chromosomes both within and between varieties as well as between different cells of individual maize plants could not be sustained in this investigation.

Therefore, his conclusion that sweet varieties are usually characterized by having twelve chromosomes, and starchy varieties by having ten, as the haploid number could not be verified in *Zea mays* var. everta. Also, Humphrey (1933) reported a counts of 2n = 22 and 2n = 26 in some varieties of *Zea mays*. But in the present investigation in var. everta no deviations in numerical status were recorded. The result obtained here tallies with those reported by Sadder and Webber (2001) using centromere specific probe CentC. Prominent CentC signals were reported on chromosome numbers 1 and 2 while chromosome 5 produced weak signals.

The basic general morphology of the chromosomes showed heterogeneous structure. Seventy percent of the chromosomes are non-metacentrics – ie six submetacentric pairs with one subtelocentric pair. This is confirmed by the calculated value of Total form percent of 35.28% indicating a reasonable degree of heterogeneity in karyotype characteristic. The longest chromosome (chromosome 1; 5.87 µm) was more than twice the length of the shortest (chromosome 10; 1.92 µm). Among the 3 accepted metacentrics observed in this study only chromosome 2 had exactly

median centromere (Table 1). In chromosome 7, the longarm is longer than thrice the length of the shortarm. These characteristics, coupled with the high total asymmetry (A_1 + A_2 = 0.8862) are indicative of a highly asymmetrical karyotype. These observations tend to confirm previous conclusion that interchanges have occurred at different sites involving different chromosomes within the *zea mays* var. everta karyotype Piagliarini *et al*, (2003). Though, the result of Piagliarini and his team could not identify the actual chromosomes involved in the interchanges, the result of the present investigation suggest that many of these interchanges have involved the shortarm. The coefficient of variation in the shortarm of 84.15%, a value which indicates high degree of variability suggests a reasonable level of both major and cryptic activities in this segment of the chromosome structure

These parameter values are indicative of the existence of genetic instability in the karyotype. The phenomenon of genetic instability occasioned by the existence of high karyotypic asymmetry and coefficient of variation have been used as premises for suggesting plant improvement programmes (Anand and Latha, 2003; Bakshi, *et al*., 2004; Dhamavanthi, 2005; Oyema *et al*., 2006; Egbucha *et al*., 2013). In their work on *Anacardium occidentale*, Aliyu and Awopetu (2007) used the result of genetic variability study on the crop to recommend a recurrent selection strategy which was largely successful and led to improvement of the crop.

CONCLUSIONS

The karyotype characteristics of *Zea mays* var. everta as revealed in this study show that the potential for improvement using regimes as simple recurrent selection exists in this plant. The high level of total asymmetry and the high degree of coefficient of variation, especially of the shortarm length is indicative of a history of both major and cryptic karyotypic activities. The variations in chromosome morphology are suggestive of genetic instability which could be exploited in further improvement.

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