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Expression of Cytological Variability in Some Miscellaneous Legume Accessions

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Abstract

Variability studies through mitotic and meiotic analyses were considered among twelve (12) species of miscellaneous legumes with two accessions of each species considered. The mitotic and meiotic procedures were carried out following an appropriate relevant literature. Generally, the somatic chromosome counts of 2n=22 were very common in the various stages observed, with the meiotic count of n=11. The mitotic metaphase chromosome were small in sizes and could be described as being metacentric and sub metacentric, often with one or two pair of satellite chromosomes. Many accessions revealed small chromosomes in most of the species that were not easily distinguished in mitotic cells, most significantly observed in the Vigna spp; Vigna radiata (green gram), Vigna mungo (mung bean) etc. and in Sphenostylis stenocarpa (African yam bean). Although, somatic chromosome counts of 2n = 22 and meiotic number of n = 11are very common among the studied accessions, there is also evidence that chromosome number different from this has been reported for miscellaneous legumes, hence there is an expression of variability in the cytological behaviors of the studied taxa.

Keywords: Mitotic; Meiotic; Variability; Chromosome; Accession

Introduction

Miscellaneous plants are the unpopular species with underutilized potentials; they are also referred to as minor, less, orphan, under cultivated, underutilized and uncommon species [1,2]. The miscellaneous legume species are the unpopular grain legumes which have received insufficient research surveillance unlike the major grain legumes like cowpea and soybean etc. The abandonment has led to the loss or genetic erosion of the germplasm (the genetic source material used by the breeders to develop new cultivars) of many of the minor legumes. Until recently, majority of them are neither cultivated nor found in the wild, they are only maintained or kept as germplasm in some research organizations or institutes for posterity. Majority of the representative

miscellaneous legumes have not witnessed reasonable research attentions over the year [3].

The use of cytological procedure to resolve evolutionary challenge has been known for ages. Chromosome number, configuration or structure as well as chromosome behavior are particularly of tremendous tool to determine variability among taxa. Data on chromosome number is a "pointer" to the role of numerical disparity while karyotypic statistics assist to apprehend the role of structural changes in the process of evolution [4]. This work was designed to carry out cytological studies through mitotic and meiotic procedures on the twelve species of the miscellaneous legumes with a view to ascertaining their somatic chromosome number and meiotic chromosome behavior. Result obtained serves as useful evaluation data to improve knowledge on their cultivation, utilization and genetic potentials.

Materials and Methods

The twelve species of the twenty four miscellaneous legumes accessions studied are: Canavalia gladiate (Sword bean-TCg1), Canavalia gladiate (Sword bean-TCg4), Pachyrhizus tuberosus (Mexican yam bean-TPtu1), Pachyrhizus tuberosus (Mexican yam bean-TPtu5), Psophocarpus tetragonolobus (Winged bean-TPt12), Psophocarpus tetragonolobus (Winged bean-TPt18), Canavalia ensiformis (Jack bean-TCc1), Canavalia ensiformis (Jack bean-TCc3), Vigna angularis (Rice bean-TVa1), Vigna angularis (Rice bean-Va1173), Vigna mungo (Mung bean-TVm12), Vigna mungo (Mung bean-TVm13), Lablab purpureus (Lablab bean-TLn21), Lablab purpureus (Lablab bean-TLn29), Sphenostylis stenocarpa (African yam-TSs137), Sphenostylis stenocarpa (African yam-TSs156), Vigna subterranean (Bambara groundnut-TVsu1126), Kerstingiella geocarpa (Kersting groundnut-TKg6), Kerstingiella geocarpa (Kersting groundnut-TKg12), Vigna radiata (Green gram-TVr45,)Vigna radiate (Green gram-TVr1001), Cajanus cajan (Pigeon pea-TCc8127), Cajanus cajan (Pigeon pea-TCc8156).

Mitotic studies

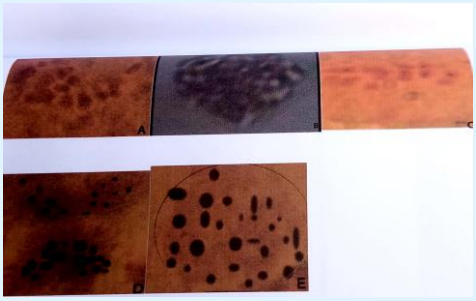
Root tips were generated from sprouted seeds (obtained from National Centre for Genetic Resources and Biotechnology (NACGRAB) Ibadan, Oyo-state, Nigeria) plated on moistened germination paper in specially made germination plastic. The root tips were pretreated with 0.04% Colchicine solution when the radicles were about 1cm long for three hours between 9am and 12 noon day to keep the majority of the cells at metaphase stage. The root tips were rinsed with tap water and fixed in 1:3 acetic acid /ethanol (v/v) for twenty four hours before use or kept in the refrigerator. The root tips were further hydrolysed with 1NHCL for 5minute and then rinsed with tap water. Slides were prepared using squashing method of Olorode [5] as modified by Adegbite and Olorode [6]. A drop of FLP – orcein stain was applied on the squashed root before applying a cover slip on each slide. Each prepared slide was mounted and observed under the microscope to search for dividing cells. Photomicrographs of good mitotic stages were taken at X 1000 magnification under oil immersion.

Meiotic Studies

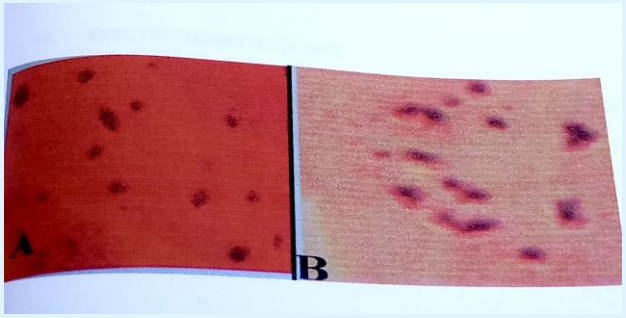
Meiotic studies were carried out on pollen mother cells obtained from young flower buds collected between the hours of 9am and 12.00 noon day. The young flower buds were fixed directly in 1:3 acetic acid/ethanol (V/V). The fixed buds were dissected to extract the young anthers. Two of the young anthers were squashed at a time on a clean slide using mounted needle and irrigated with the fixation until homogeneous solution was obtained. A drop of FLP-orcein stain was applied on the slide after which a cover slip was also applied. The slides were observed under the microscope cells. Photomicrographs of good meiotic stages were taken at X1000 magnification under oil immersion.

Results

Cytology of the various accessions and species of the miscellaneous legumes studies revealed small chromosomes in most of the species that were not easily distinguished in mitotic cells, most significantly observed in the Vigna spp.; Vigna radiata (green gram), Vigna mungo (mung bean) etc. and in Sphenostylis stenocarpa (African yam bean). In the majority, the somatic chromosome counts of 2n=22 were very common in the various stages observed, with the meiotic count of n=11. The mitotic metaphase chromosome were small in sizes and could be described as being metacentric and submetacentric, often with one or two pair of satellite chromosomes. Chromosome were appeared to be interwoven and coiled at the early prophase stages and could not be counted while there were appearances of chromatin shortening to chromosomes at the late prophase stage. The metaphase chromosomes were clearly distinguishable and were observed to have aligned themselves at the metaphase plate. Figures 1 & 2 present the appearance of the various stages observed.



A. Pre-metaphase stage in TSs 137 (2n=22)
B. Early Prophase stage in TPt 12
C. Late Metaphase stage in TCg 4 (2n=22)
D. Anaphase stage in TVm 12
E. Pre-metaphase in TVr 1001
Figure 1: Appearance of mitotic chromosomes in selected miscellaneous legumes studied



A. Metaphase 1 stage in TVsu 1126 (n=11)
B. Metaphase 1 stage in TSs 137 (n=11).
Figure 2: Appearance of meiotic chromosomes in selected miscellaneous legumes studied.

Discussion

In all the glaringly different species of the miscellaneous legumes, Sphenostylis stenocarpa, Psophocarpus tetregonelobus, canavalia gladiata, Vigna mango and Vigna radiata (TSs137, Tpt12, TCg4, TVm12 and TVr1001 respectively) considered in this study , a somatic chromosome counts of 2n=22 and meiotic number of n =1 were made in some cells. The chromosomes were small, mostly metacentrics and submetacentrics with one or two pairs of satellite chromosomes. This is similar to previous studies reported for miscellaneous legumes. For example, Peter & Davidse, 1977 [7] (for the genus Sphenostylis and a sister spieces of Sphenostylis marginata); Munthas, et al. [8] (for Vigna radiata); Brink, et al. [9] (for Vigna subterannea), however, mitoitic chromosome counts of 2n =20 and 2n=18 were also observed in previous studies for Kerstingiella-geocarpa Dako & Vodouhe [10] and Psophocarpus tetragonolobus Grubben [11] respectively.

Conclusion

The different species of the miscellaneous legumes considered in this study had somatic chromosome counts of 2n = 22 and meiotic number of n = 11. This further lends credence to their genetic relatedness. However, mitotic/somatic chromosome count different from this has also been reported for miscellaneous legumes (Dako and Vodouhe, 2006). We conclude that there is expression of variability in the cytological characters of the studied taxa.

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