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Mitotic Studies on *Combretum* Loefl. from Nigeria

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ABSTRACT

Cytological studies involving root-tip chromosomes of ten *Combretum* species belonging to the family Combretaceae from Nigeria were carried out. The result showed that *C. platypterum*, *C. racemosum*, *C. constrictum*, *C. bracteatum and a yet to be identified Combretum* sp.2 have a chromosome number of 2n = 26. Also, *C. racemosum*, *C. zenkeri* and *Combretum* sp.3 (*yet to be identified*) have a chromosome number of 2n = 39. A basic chromosome number of x = 13 for the genus is therefore proposed.

Keywords: *Combretum*; Combretaceae; Cytology; Mitotic Chromosomes

1. Introduction

Combretaceae is family of plants with 20 genera and 600 species of tropical and subtropical regions of the world [1]. In West Africa, the family Combretaceae is represented by 9 genera with 72 species and the genus *Combretum* Loefl. is the largest genus with 48 species and 8 imperfectly known species [2]. In Nigeria, the genus *Combretum* Loefl. is represented by 25 species which are mainly straggling shrubs or lianes [3]. The acid extracts from the mature leaves of *C. bracteosum* are used as mild steel corrosion inhibitor [4] while some other members of the genus produce gum exudates of high viscosity and high molecular weight, usable in bio-engineering [5,6]. Numerous ethnopharmacological, anti-oxidant and antimicrobial characteristics have been reported for members of the genus [7-10]. Despite the importance of members of this genus, there is paucity of cytotaxonomic information regarding the species.

The chromosome number as one of the three headings of cytotaxonomy is of great importance in taxonomic studies. Stace [11] stated that "chromosome number and homology largely determines pairing behaviour at mitosis, which in part govern the level of fertility of hybrids and hence the breeding behaviour and pattern of variation of populations". Apart from reports of basic number of x = 12 and 13 [12] for the family, there is no known report on the chromosome numbers of members of this genus in Nigeria. From the work of Stace [11], it is obvious that knowledge of the chromosome number is important in the manipulation and improvement of plant species. This and the need to improve on the taxonomy of the genus underscore this study on a genus with great horticultural importance. The findings of this study represent the first known report of chromosome numbers of *Combretum* species in Nigeria.

2. Materials and Methods

Plant specimens were collected from various parts of South-South and South-Eastern Nigeria as shown in **Table 1**. These specimens were taken to Forestry Herbarium Ibadan (FHI) and properly identified by comparing with deposited materials in the herbarium and reference to the Flora of West Tropical Africa [2].

Cytological examinations were carried out on root tips generated from the specimens collected between 8 a.m. and 10 a.m. The young root tips were pretreated with 0.02 M 8-hydroxyquinoline for 3 to 4 hr, fixed in Carnoy's fluid (glacial acetic acid and ethanol, 1:3 ratio), transferred to 70% ethanol and stored in a refrigerator till required. Thereafter, the specimens were removed, placed in 18% hydrochloric acid (HCl) for 2 minutes, squashed using 2% FLP Orcein, viewed under a Meiji microscope (MX 4200L) and chromosome number determined [13]. Measurement of chromosome lengths was achieved with an ocular (X10) fitted with a micrometer rule and good chromosome plates photographed with Canon A480 digital camera fitted on Leitz Diaplan microscope. For each species, ten chromosomes were randomly measured from five different preparations. The lengths of the chromosomes were summed up for each species; the mean lengths and standard deviations thereof were statistically computed in EXCEL.

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Table 1. Species name, locality, co-ordinates and dates of collection of *Combretum* species studied.

S/N	Species name	Locality	Co-ordinate/elevation	Date of collection
1	*C. bracteatum* (Laws.) Engl. & Diels.	Along Aba Port Harcourt express way, by Asa High Secondary School, Abia State	N04 55' 45.40", E007 14' 28.70"/34 m	February 5, 2011
2	*C. constrictum* (Benth.) Laws	Bank of Taylor Creek, Zarama Bayelse State	N05 10' 55.20", E006 13' 36.20"/23 m	December 12, 2010
3	*C. dolichopetalum* Engl. & Diels.	Elikpokwuodu/Mgbuchi road by Obio-Akpor Modern Market, Port Harcourt, Rivers State	N04 53' 43.00", E006 59' 49.00"/30 m	May 2, 2011.
4	*Combretum* sp. 3	Ogboinbiri, Bayelsa State	N04 43' 08.80", E005 57' 06.70"/24 m	November 20, 2010
5	*C. hispidum* Laws	This species is located behind Total Fuel Station East/West road Choba, Port Harcourt	N04 53' 24.80", E006 54' 59.80''/19 m	January 8, 2011.
6	*C. mooreanum* Exell.	UPTH road, Alakahia, Rivers State	N04 58' 24.40", E006 55' 35.00"/17m	December 31, 2010
7	*C. platypterum* (Welw.) Hutch. & Dalz.	Opposite Choba Police Station, along East/West road, Port Harcourt	N04 53' 32.20", E006 54' 57.20"/16 m.	September 4, 2010.
8	*C. racemosum* P. Beauv.	Beside Choba borrow pit along East/West road, Rivers State	N04 53' 24.20", E006 55' 09.40"/18 m	December 31, 2011.
9	*C. zenkeri* Engl. & Diels.	Mgbirichi Community along Owerri-Port Harcourt Road, by Hausa settlement, Imo State	N05 21' 49.20", E006 57' 30.00"/44 m	August 25, 2010/ September 10, 2010/ May 8, 2011.
10	*Combretum* sp.1 Ak	Ikot Ekwere, Akwa Ibom State	N04 45' 46.80", E007 59' 55.60"/17 m	August 30, 2011

3. Results

The cytological observations on the taxa are presented in **Table 2** and **Figure 1(A)-(J)**. *C. mooreanum*, *C. platypterum*, *C. bracteatum*, *C. hispidum*, *C. dolichopetalum* and *Combretum* sp.2 have somatic chromosome number of 2n = 26. In *C. racemosum* and *Combretum* sp.3, a somatic chromosome number of 2n = 39 was recorded. However, variable somatic chromosome numbers of 2n = 26 and 39 were observed for *C. zenkeri.* These results suggest a basic number of x = 13 for the species studied.

The sizes of the chromosomes varied from species to species (**Table 3**). For instance, the smallest chromosome length was observed in *Combretum* sp.3 (29.4 ± 9.78 µm) while the longest were observed in *C. mooreanum* (84.65 ± 17.13 µm). The lengths of chromosomes in the other species studied included *C. hispidum* (40.38 ± 6.73 µm), *C. constrictum* (39.42 ± 10.31 µm), *C. bracteatum* (44.91 ± 12.58 µm), *Combretum* sp.2 (29.08 ± 8.86 µm), *C. racemosum* (51.69 ± 14.03 µm), *C. zenkeri* (56.86 ± 18.9 µm), *C. platypterum* (41.35 ± 8.40 µm) and *C. dolichopetalum* (41.68 ± 6.57 µm).

4. Discussion

Existing information on the chromosome numbers of this genus is scanty. Brighton and Wickens [12] reported a basic chromosome number of x = 12 or 13 in Combretaceae. In the genus *Combretum*, they reported basic numbers of x = 13 and 2n = 26, 39, 52, 78 and 104 suggestive of diploid, triploid, tetraploid, hexaploid and octoploid levels. In this present study, ten species of the genus showed different cytotypes. *C. mooreanum*, *C. platypterum*, *C. bracteatum*, *C. hispidum*, *C. dolichopetalum*, *C. consrtictum* and *Combretum* sp.2 are diploids with chromosome numbers of 2n = 2x = 26. However, *C. racemosum* and *Combretum* sp.3 are triploid. Their somatic chromosome number is 2n = 3x = 39. On the other hand, *C. zenkeri* is a mixoploid having chromosome number of 2n = 2x = 26 (diploid) and 2n = 3x = 39 (triploid) occurring in the somatic cell. This study confirms basic chromosome number of x = 13 for the *Combretum* species studied and is in agreement with earlier observations [12].

Polyploidy plays important role in the speciation of higher plants [11]. This occasionally manifests morphologically in variations such as flower colour, leaf and fruit sizes [14] with larger leaf sizes and reduced fertility (especially in triploids) being common with polyploids against their diploid counterparts [14,15]. Diploid members of *Combretum* studied were observed to have large broad leaves with showy inflorescences and large fruits as against the triploids, with reduced lance-shaped leaves. Inflorescences and fruits in the triploids were also reduced in size. Though these agree with earlier reports [14-16], further studies (field and experimental) are on-

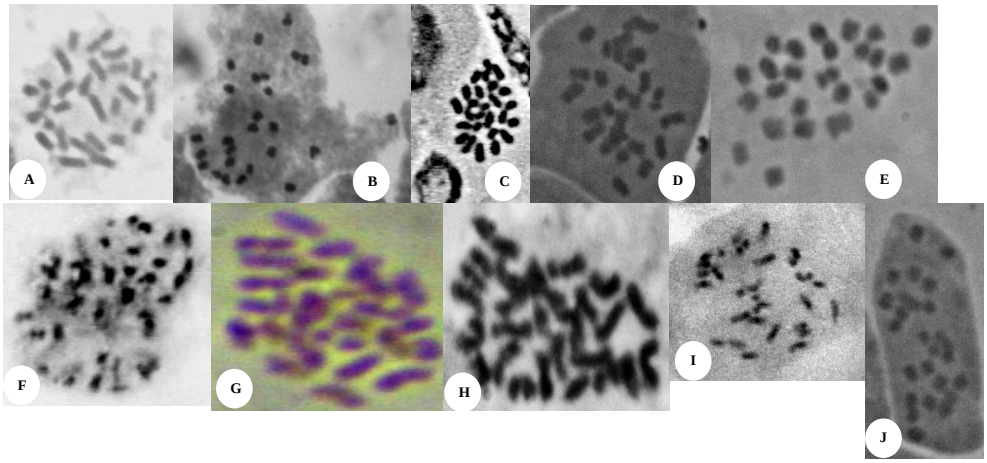

Figure 1. Chromosome configuration in the *Combretum* species studied. A. *C. mooreanum* (2n = 26); B. *Combretum* sp.2 (2n = 26); C. *C. platypterum* (2n = 26); D. *C. bracteatum* (2n = 26); E. *C. hispidum* (2n = 26); F. *Combretum* sp.3 (2n = 39); G. *C. zenkeri* (2n = 39); H. *C. racemosum* (2n = 39); I. *C. dolichopetalum* (2n = 26); and J. *C. constrictum* (2n = 26).

Table 2. Chromosome numbers of species studied in the genus *Combretum*.

S/N	Species name	Chromosome number
1	*C. platypterum*	2n = 2x = 26 (diploid)
2	*C. racemosum*	2n = 2x = 39, x = 13 (Triploid)
3	*Combretum* sp.2	2n = 2x = 26, x = 13 (diploid)
4	*Combretum* sp.3	2n = 2x = 39, x = 13 (Triploid)
5	*C. dolichopetalum*	2n = 2x = 26, x = 13 (diploid)
6	*C. hispidum*	2n = 2x = 26, x = 13 (diploid)
7	*C. zenkeri*	2n = 2x = 26, x = 13 (diploid) 2n = 3x = 39, x = 13 (Triploid)
8	*C. bracteatum*	2n = 2x = 26, x = 13 (diploid)
9	*C. mooreanum*	2n = 2x = 26, x = 13 (diploid)
10	*C. constrictum*	2n = 2x = 26, x = 13 (diploid)

Table 3. Lengths of chromosomes in the *Combretum* species studied.

Species name	*Mean ± STD (µm)	Range (µm)	Coefficient of variation
C. hispidum	40.38 ± 6.73	33.6 - 50.4	16.67
C. constrictum	39.42 ± 10.31	21.0 - 63.0	26.15
C. bracteatum	44.91 ± 12.58	21.0 - 63.0	28.01
Combretum sp.2	29.08 ± 8.86	16.8 - 42.0	30.46
C. mooreanum	84.65± 17.13	105.0 - 42.0	20.23
C. racemosum	51.69 ± 14.03	25.2 - 84.0	27.13
C. zenkeri	56.86 ± 18.90	33.6 - 84.0	33.23
C. dolichopetalum	41.68 ± 6.57	33.6 - 54.6	15.76
C. platypterum	41.35 ± 8.40	25.2 - 54.6	20.31
Combretum sp.3	29.40 ± 9.78	16.8 - 42.0	33.27

*Measurements based on 16 cells.

going for detailed morphological and reproductive comparison of features between the diploids and polyploids in the genus.

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