

Full Length Research Paper

Chromosome number and cytomorphological characterization of a polyploid *Abrus*

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Accepted 16 March, 2005

Chromosome counts from natural populations of *Abrus pulchellus* in Nigeria were carried out. Tetraploid ($2n = 44$) chromosome number was constant in all the samples investigated. The 44 chromosomes fall into three cytomorphological categories: eight metacentric and eight sub-metacentric pairs, and six acrocentric pairs. The chromosomes are relatively small in length ranging from 0.5 to 1.4 μ m. The polyploid (tetraploid) cytotype is reported for the first time for this taxon.

Key words: *Abrus pulchellus*, acrocentric, chromosome counts, metacentric, polyploid, sub-metacentric.

INTRODUCTION

The genus *Abrus* Adanson is a small pantropic genus (Isely, 1990) of the tribe Viceae of Papilionoidae-Leguminosae (Hutchinson and Dalziel, 1958). In West Africa, Hutchinson and Dalziel (1958) reported three species: *A. precatorius* Linn, *A. pulchellus* Wall. and *A. canescens* Welw. ex Bak., all of which occur in Nigeria. At present *A. canescens*, which used to occur in the northern savanna area of Nigeria is almost extinct (Agbagwa, 2001), thus increasing the need for more studies on the genus. Though *Abrus* members are lesser known and utilized among the legumes, great potentials for exploitation abound for the species. Some species have been employed in ethnomedical, ethnopharmacological and toxicological uses (Dalziel, 1937; El-Shabrawy et al., 1987; Amer et al., 1989; Reda et al., 1989; Dimetry et al., 1990; Sinha, 1990; Burkill, 1995). The leaves and leafy twigs of some are used in the preparation of liquorice (Choi et al., 1989; Jakinovich et al., 1990).

Chromosome numbers have been reported for *A. precatorius* (Borgen, 1980; Gill and Husaini, 1986; Yeh et al., 1986; Kumari and Bir, 1990; Agbagwa, 2001). While Gill and Husaini (1986) gave a gametophytic count of 11

for *A. precatorius*, the other authors expectedly reported sporophytic count of 22 for the same species. The existing chromosome information encountered before this study for *A. pulchellus* are the gametophytic counts of 12 by Gill and Husaini (1986) and Chatterjee (1989). So far polyploidy has not been reported in the genus. Agbagwa (2001) carried out a biosystematic study on *Abrus* in Nigeria and observed paucity of information on the cytology of the taxa. Added to this is the rather confusing basic chromosome number of 11 and 12 for the few reports on the genus, which hitherto suggests aneuploidy. However, the consistent 44 chromosomes observed in this study across natural populations of *A. pulchellus* in Nigeria points to a tetraploid status based on $2n=22$ for *A. precatorius* and agrees with a basic chromosome number of 11.

MATERIALS AND METHODS

Root tips for mitotic studies were obtained from seeds planted after scarification in buckets containing white sand. The time of collection of root tip was between 11 am and 12 noon. The roots were washed and excised using a pair of forceps and pretreated with 0.002 M 8-hydroxyquinoline solution for 3 h. Fixation of the rootstock in acetic ethanol (Clarke's fluid) followed the method of Okoli (1983). The rootstock were preserved after fixation in 70% (v/v) ethanol solution and stored in a refrigerator till required for squashing.

In order to observe mitotic chromosomes, root tips preserved in 70% ethanol solution were hydrolyzed in 18% HCl aqueous for 3

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Figure 1. Mitotic chromosomes of *Abrus puchellus*. 2n=44 x 400.

Table 1. Localities from which *A. pulchellus* seed samples were collected for the mitotic counts, chromosome numbers and sizes.

Collection number	Locality	State/District	Collector	2n	Size in μm	Geographical coordinates
001	Emeabiam	Owerri – West, Imo State	Agbagwa 003	44	0.5 – 1.10	05° 29" N, 07° 05"E
002	University of Port Harcourt		Agbagwa 004	44	0.5 -1.40	05° 45" N, 07° 15"E
003	Reserved forest, University of Ibadan	Ibadan, Oyo	Agbagwa 006	44	0.5 – 1.20	07° 23" N, 03° 54" E
004	High rainforest, Okpai/Kwale	Ndokwa East, Delta State	Agbagwa 007	44	0.5 – 1.40	05° 42" N, 06° 29" E
005	KM 4, along Kolo-Creek/Rumuekpe Pipeline, Imiringi,	Bayelsa State	Agbagwa 009	44	0.5 – 1.10	04° 54" N, 06° 22"E
006	High Secondary forest, opposite IITA office, Onne Station	Rivers State	Agbagwa 012	44	0.5 – 1.10	04° 51" N, 07° 03"E



Figure 2. Karyotype of the 44 Chromosomes of *A. Pulchellus*.

min. Hydrolyzed root tips were rinsed quickly in 70% ethanol. The root tip (about 1 mm long) was excised and squashed in a drop of FLP-Orcein (a solution of Orcein and equal portion of formic, lactic and propionic acids in water; Okoli, 1992) under No.0 or No.1 cover glass. The cells were flattened-out by pressing firmly with the thumb. The temporary mounts were observed under Leitz light microscope at X40 objective lens.

Chromosome counts and description were carried out using the oil immersion objective. Measurements of chromosome lengths was achieved with an ocular (X10) fitted with a micrometer rule and good chromosome plates photographed with Leica WILD MPS 52 microscope camera on Leitz Diaplan microscope.

RESULTS

A chromosome number of $2n=44$ (Figure 1) was recorded in all six populations of *A. pulchellus* used for this study (Table 1). The results show that the size of the chromosomes ranges from 0.5 to 1.4 μm in each sample. A painstaking karyotype analysis of the 44 chromosomes (Figure 2) revealed three morphological categories: eight metacentric pairs, eight submetacentric pairs and six acrocentric pairs. The pairing is as follows: metacentric pairs are chromosomes 3 and 4, 5 and 6, 13 and 14, 17 and 18, 19 and 20, 31 and 32, 37 and 38, 43 and 44; acrocentric pairs are 23 and 24, 25 and 26, 27 and 28, 29 and 30, 33 and 34, 39 and 40; while submetacentrics are 1 and 2, 7 and 8, 9 and 10, 11 and 12, 15 and 16, 21 and 22, 35 and 36, 41 and 42. On the basis of size, the 44 mitotic chromosomes of *A. pulchellus* can be grouped into three. Chromosomes 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11 and 12 are large; eight other chromosomes (37, 38, 39, 40, 41, 42, 43 and 44) are small while 24 chromosomes (i.e. from chromosome 19 to 36) are intermediate. Among the intermediate group, a further distinction into two groups may be made on the basis of size. Chromosomes 15, 16, 23, 24, 27, 28, 35 and 36 constitute the smallest group. Between the remaining 16 pairs of chromosomes of the intermediate group and the large chromosome group, there seems to be a continuous variation in size. Thus, there was occasional difficulty in distinguishing them based on size. A further distinction based on size could be done among the large group. Chromosomes 1, 2, 5 and 6 are the largest and form their own group among the large ones. Telocentric chromosomes were not observed.

DISCUSSION

The chromosome number of $2n=44$ found in all samples of *A. pulchellus* in the present study is the first report for this species. This does not agree with the sparse literature on this species. Gill and Husaini (1986) and Chatterjee (1989) reported gametophytic count of $n=12$ for this taxon, which implies $2n=24$. Gametophytic count of 11 (Gill and Husaini, 1986) and sporophytic count of 22 (Borgen, 1980; Yeh et al., 1986; Kumari and Bir, 1990;

Agbagwa, 2001) have been made for *A. precatorius*, another *Abrus* species. The results of this present study simply suggest a doubling of the 22 chromosomes of *A. precatorius* to arrive at the 44 in *A. pulchellus*. Thus, *A. pulchellus* is likely a polyploid (tetraploid) with *A. precatorius* as one of the probable diploid progenitors. The basic chromosome number for the two species based on the foregoing is $x=11$.

We do not rule out the possibility of aneuploidy in the genus as being suggested by gametophytic counts of 11 and 12 earlier reported for *A. precatorius* and *A. pulchellus* respectively, since this is common in some genera of the Leguminosae (Sanjappa and Bhatt, 1977, 1979; Gupta and Gupta, 1978; Coleman and Demenezes, 1980; Polhill and Raven, 1981; Gill and Husaini, 1985, 1986; Ugborogho and Obute, 1990). However, our chromosome counts from different populations of this same species from different localities/areas in Nigeria (Table 1) convince us that the exact chromosome number for *A. pulchellus* is 44. It is possible that in the earlier reports there was confusion in species identity, for there exists in Nigeria a third species of *Abrus* (*A. canescens*) (Hutchinson and Dalziel, 1958). *A. canescens*, according to Agbagwa (2001), is presently rare in occurrence if not extinct. Hutchinson and Dalziel (1958) in describing *A. pulchellus*, made reference to an earlier use of the name *A. canescens* for *A. pulchellus* either as a synonym or outright wrong naming. Agbagwa (2001) also observed that the floral description of *A. precatorius* and *A. pulchellus* made in Hutchinson and Dalziel's Flora of West Tropical Africa (1958) did not match with collections made in Nigeria between 1998 and 2002. These facts only suggest that there could be a mistaken identity of species in earlier chromosome counts.

Observations on chromosome morphology and size made in this study are an insight into what will be expected at meiosis. Distribution of several morphological categories namely acrocentrics, metacentrics and submetacentrics occurring in different size regimes as was observed, is likely to result in many structural changes and rearrangements at meiosis, which could lead to speciation. Akpabio and Olorode (1988) observed similar trend in *Crotalaria*, another leguminous genus.

REFERENCES

- Agbagwa IO (2001). Biosystematic Studies on the Genus *Abrus* Adanson in Nigeria; Ph.D. Thesis, Univ. of Port Harcourt, Port Harcourt, Nigeria.
- Akpabio KE, Olorode O (1988). Morphological and Karyotype studies in eight species of the genus *Crotalaria* Linn. Nig. J. Bot. 1: 21-35.
- Amer SA, Reda AS, Dimetry NZ (1989). Activity of *Aburs precatorius* L. extracts against the two-spotted spider mite *Tetranychus urticae* Koch. Acarologia 30: 3, 209-215.
- Borgen L (1980). Chromosome numbers of Macaronesian flowering plants III, Botanica Macaronesia 7: 67-76
- Burkill HM (1995). The Useful Plants of West Tropical Africa. (Ed. 2), Vol. 3. Families J-L. Roy. Bot. Gardens Kew.

- Chatterjee A, Ghosh S, Roy SC (1989). A cytological survey of eastern Himalayan plants III. Cell Chromosome Res. 12: 22-29.
- Choi YH, Hussain RA, Pezzuto JM, Kinghorn AD, Morton JF (1989). Abrusosides A-D, four novel sweet-tasting triterpene glycosides from the leaves of *Abrus precatorius*. J. Nat. Prod. 52:5, 1118-1127.
- Coleman JR, Demenezes EM (1980). Chromosome numbers in Leguminosae from state of Sao Paulo, Brazil. Rhodora 82: 475 – 481.
- Dalziel JM (1937). The Useful Plants of West Tropical Africa, ed. 1, Crown Agents for the Colonies, Lond..
- Dimetry NZ, El-Gengaihi S, Reda AS, Amer, SAA (1990). Toxicity of some compounds isolated from *Abrus precatorius* L. seeds towards the two -spotted spider mite *Tetranychus urticae* Koch. Acarologia 31:4, 361-366.
- El-Shabrawy OA, El-Gengaihi S, Ibrahim NA, Ali-Ibrahim N (1987). Toxicity and teratogenicity of abrin. Egypt. J. Vet. Sci., 24:2,135-142.
- Gill LS, Husaini SW (1985). Cytological evolution of the Southern Nigeria Leguminosae. Rev. Cytol. Biol. Veg. Bot. 8:3-31
- Gill LS, Husaini SW (1986). Cytological observations in Leguminosae from Southern Nigeria. Willdenowia 15:521-527.
- Gupta R , Gupta PK (1978). Karyotype studies in the genus *Crotalaria* L. Cytologia 43: 357-369.
- Hutchinson J , Dalziel JM (1958). Flora of West Tropical Africa, Crown Agents, ed. 2. (Revised by R. W. J. Keay), pp. 574-575.
- Isely D (1990). Vascular flora of the Southeastern U.S.A Vol. 3, Part 2 Leguminosae (Fabaceae). Univ. of North Carolina Press, Chapel Hill, NC. p. 277.
- Jakinovich W, Moon C, Choi YH , Kinghorn AD (1990). Evaluation of Plant extracts for sweetness using the Mongolian gerbil. J. Nat. Prod. 53:1, 190-195.
- Kumari S, Bir SS (1990). Karyomorphological evolution in Papilionaceae. J. Cytol. Genet.. 25:173-219.
- Okoli BE (1992). Field, Herbarium and Laboratory Techniques. Mbeyi and Associates (Nig.) Ltd., Port Harcourt, 239 p.
- Okoli BE (1983). Hybridisation, Polyploidy and Apomixis in *Andropogon tectorum*, Schum and Thorn (Gramineae). New Phytol. 93: 591-597.
- Polhill RM, Raven PH (1981). Advances in Legume Systematics, Part 1. Royal Botanic Gardens, Kew, England.
- Reda AS, Dimetry NZ, Amer SA, Motawe, HM (1989). Activity of *Abrus precatorius* L. extracts and compounds isolated on orientation and oviposition behaviour of the two-spotted spider mite *Tetranychus urticae* Koch. J. Appl. Entomol. 107(4): 395-400.
- Sanjappa M, Bhatt RP (1977). In 10PB Chromosome Reports L111. Taxonomy 25:483-500.
- Sanjappa M , Bhatt RP (1979). Cytological observations in the family Fabaceae: tribe Indigofereae. J. Cytol. Genet. 14: 58-63.
- Sinha R (1990). Post testicular antifertility effects of *Abrus precatorius* seed extract in albino rats. J. Ethnopharmacol. 28(2):173-181.
- Ugborogho RE, Obute GC (1990). Cytomorphological consideration of *Vigna unguiculata* (L) Walpers. species complex (*Papilionaceae*) in Southern Nig. Bull. Sci. Assoc. Niger. 16 (1):91.
- Yeh MS, Yuasa H ,I Maekawa F (1986). Chromosome numbers in the Leguminosae. Sci. Rep. Res. Inst. Evol. Biol. 3:57-71.