A Cytogenetical Study of Intergeneric Hybrids Between *Bothriochloa intermedia* and *Dichanthium fecundum*

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The occurrence of natural interspecific and intergeneric hybrids is perhaps more frequent in the Gramineae than any other family of flowering plants. Studies of these natural, as well as artificially produced, hybrids and their progenies have not only helped to increase our understanding in the fields of genetics and cytogenetics but have also helped the taxonomist to understand the relationships between various taxa from a biosystematic and phylogenetic point of view. These studies often provide the plant breeder important information in his efforts towards the development of a better adapted crop plant.

Myers (1947) reported about 225 interspecific and intergeneric hybrids in forage grasses, and following him Carnahan and Hill (1961) listed 226 interspecific and 95 intergeneric hybrids. The number of such studies is steadily increasing. In the generic group Bothriochloinae, subtribe Andropogoninae, hybrids within and between the genera, *Bothriochloa* O. Kuntze *Capillipedium* Stapf. and *Dichanthium* Willemet have been reported (Harlan *et al.* 1958, 1961, Celarier *et al.* 1961, de Wet *et al.* 1961, and Chheda *et al.* 1961). Borgaonkar and de Wet (1960) from their studies of hybrids between *Dichanthium annulatum* and *D. fecundum* and a backcross population of the hybrid with *D. annulatum*, suggested that the sexuality of pedicellate spikelet, a basic difference between the two species, is controlled by a single gene.

The present paper refers to a cytogenetical study of intergeneric hybrids between *Bothriochloa intermedia* (R. Br.) A. Camus and *Dichanthium fecundum* S. T. Blake.

The highly polymorphic species of *Bothriochloa intermedia* extends from Southern Africa through the tropics and subtropics to Australia and adjacent islands. It appears to introgress with other species of *Bothriochloa*, *Dichanthium* and *Capillipedium* where they are sympatric (Harlan *et al.* 1961). Apomixis, however, has always been a strong barrier to the production of hybrids within and between species of *Bothriochloa* and *Dichanthium*. By crossing two facultatively apomictic strains of the gangetica type of *B. intermedia*, obtained from India, a highly self-sterile and sexual plant X-750 was obtained. *B. intermedia* gangetica type as described by Harlan *et al.* (1961)

differs from the more typical members of the species in having subequal pedicellate and sessile spikelets, solid pedicels, some bulbous based hairs at the tip of the lower glume of both spikelets and has hair all over the nodes. On the other hand, *Dichanthium fecundum* is widely distributed in Australia but does not extend to Asia or Africa. It has sometimes been confused with *D. annulatum*, a species as widely distributed as *B. intermedia*. However, *D. fecundum* differs from *D. annulatum* in having stouter and fewer racemes with much longer spikelets. The pedicellate spikelets of *D. fecundum* are hermaphroditic a characteristic unique to only this species in the genus *Dichanthium*.

Material and methods

Hybrids were obtained by using *B. intermedia* X-750 as the female parent and *D. fecundum* 6525, obtained from Queensland, Australia, as the male parent. Reciprocal hybrids were not possible since *D. fecundum* 6525 is an obligate apomict. The parents and hybrids were grown in a uniform nursery as described by Celarier and Harlan (1956).

Morphological studies were made from herbarium specimens filed with the Oklahoma State University. Bud material for cytological studies was fixed in Carnoy's solution and stored in a refrigerator. Microsporocytes were smeared and stained with acetocarmine.

Experimental results

The distinguishing morphological characteristics of the parents and hybrids are presented in Table 1 and Figs. 1 to 5.

Table 1. Distinguishing morphological characters of the parents and hybrids

	Length of main axis	Length of longest raceme	No. of nodes	No. of racemes	Hairs at nodes	Sessile spikelet L/W	Ped. spikelet L/W	Ped. spikelet sexuality
Parents								
<i>B. intermedia</i> X-750	56 mm	67 mm	7	17	+++	3.5/1.0mm	4.0/1.1mm	Unisexual
<i>D. fecundum</i> 6525	11	94	2.4	3.7	+	5.0/1.3	5.5/1.7	Bisexual
Hybrids								
-1	34.5	79	4.5	11	++	4.6/1.0	5.3/1.5	Unisexual
-2	25.5	66	4	7	+	4.0/1.0	5.2/1.4	Bisexual
-3	31.8	69	5.3	9.7	+	4.0/1.1	4.6/1.5	Bisexual
-4	33.7	75	5.0	10.7	+	4.0/1.0	5.2/1.5	Bisexual
-5	21	62	3.5	6.5	+	4.5/1.1	5.5/1.5	Unisexual
-6	34	101	5.5	10	+	4.5/1.0	5.5/1.5	Unisexual
-7	40	100	5.3	9.3	+	4.5/1.1	5.5/1.4	Unisexual

Seven hybrids were studied in detail. From the Table and Figures it becomes apparent that the hybrids show a rather wide range of morphological

variation; however, they are generally intermediate in most characters. In contrast to the results obtained in *D. annulatum* $\times$ *D. fecundum* by Borgaonkar and de Wet (1960) where all hybrids had bisexual pedicellate spikelets, in

Figs. 1-4. Inflorescence of the parents and some hybrids. 1, $\times$ -750. 2, an unisexual hybrid (no. 6). 3, a bisexual hybrid (no. 3) and 4, 6525.

this study three of the F_1 hybrids have bisexual pedicellate spikelets and four have unisexual pedicellate spikelets. The hybrids were generally healthy and grew vigorously; some were completely sterile, others set a few seeds

Fig. 5. Scatter diagram showing some distinguishing morphological characteristics of the parents and 7 hybrids.

while one hybrid (No. 7) produced abundant seeds when left to open pollinate in the field. On self pollination the hybrids were either completely sterile or set an extremely small number of seeds.

The somatic chromosome numbers for each of these plants are presented in Table 2 together with the range and average frequency of chromosome configurations per cell at diakinesis and metaphase I. Two of the hybrids are aneuploids with $2n$ chromosome numbers of 38 and 39 and the meiotic behavior varies from plant to plant.

Table 2. Chromosome configuration of the parents and hybrids at diakinesis and metaphase I

Plant	Range					Av. per cell			
	$2n$	I	II	III	IV	I	II	III	IV
Parents									
<i>B. intermedia</i> X-750	40	0-16	12-20	0-1	0	7.30	16.30	0.03	0
<i>D. fecundum</i> 6525	40	0-3	15-20	0-1	0-2	0.46	18.42	0.03	0.65
Hybrids									
-1	40	8-16	11-16	0-1	0-1	12.30	13.33	0.15	0.15
-2	40	4-10	12-18	0-1	0-1	8.29	14.87	0.28	0.28
-3	38	16-22	8-11	0	0-1	19.27	9.18	0.00	0.09
-4	39	19-29	5-10	0	0	23.33	7.83	0	0
-5	40	11-20	8-13	0-2	0-2	15.41	11.00	0.42	0.33
-6	40	4-14	13-17	0-1	0-2	8.66	14.00	0.13	0.75
-7	40	4-14	12-18	0-1	0-1	10.6	13.82	0.25	0.25

The female parent *B. intermedia* X-750 has more irregular meiotic behavior than *D. fecundum* 6525 and has frequently produced aneuploid progeny in crosses (Chheda *et al.* 1961). *D. fecundum* 6525 is an apomictic tetraploid. Its meiotic behavior is fairly regular except that it forms an average of 0.65 quadrivalents per cell. Anaphase I, telophase I, and the second divisions are usually regular.

All the hybrids exhibit a comparatively irregular meiotic behavior. Out of the three hybrids which showed bisexuality in their pedicellate spikelets two are aneuploids with more irregular meiotic behavior than the other hybrids. The other hybrids show averages of between 8 and 15 univalents per cell. At anaphase I and telophase I, many dividing and some nondividing laggards are seen corresponding more or less directly with the number of univalents observed at metaphase. These univalents form up to six micronuclei at the dyad spore stage. The second division is also irregular and at tetrad spore stage up to five micronuclei have been recorded.

Discussion

The female parent, *B. intermedia* X-750 has been given the genomic

constitution BBDD (Harlan *et al.* 1961, Chheda 1962). Except for the character of bisexual pedicellate spikelets, *D. fecundum* has no character that is not found in other species in the genus. It may, therefore, be assigned the genomic constitution $D_rD_rD_r'D_r'$. The hybrids, then, become: BDD_rD_r' .

In other studies, *B. intermedia* X-750 has been shown to be heterozygous for a gene controlling bivalent formation (Chheda 1962). When the dominant allele is absent, only homologous pairing takes place; when it is present, homoeologous and even nonhomologous pairing occurs. In this study the data are not so clear-cut as in some others due to aneuploidy in hybrids 3 and 4, but the low frequency of pairing in these two plants suggests that the gene for compulsive pairing is not operating. If this is true, there is approximate homology between two sets of 10 chromosomes. By the notation above, the D and D_r sets must be fairly homologous while the B and D_r' sets are nonhomologous. In the other hybrids, pairing is increased under genetic control.

The aneuploidy is almost certainly due to the irregular meiosis of X-750, and the missing chromosomes are B or D, but not D_r or D_r' (Chheda *et al.* 1961).

In another study reported by Borgaonkar and de Wet (1960), the hybrids between *D. annulatum* $\times$ *D. fecundum* all had bisexual pedicellate spikelets, and in the backcross to *D. annulatum*, a 1:1 ratio was obtained. In this cross, only three of the hybrids had bisexual pedicellate spikelets, while four had unisexual pedicellate spikelets. The D genome in gangetica plants has been shown to be closely homologous to genomes in *D. annulatum* (Harlan *et al.* 1961). Therefore, there is probably a gene in the B genome which prevents the development of ovaries in pedicellate spikelets. The gene is not present in *D. annulatum*, but may represent part of a strong tendency in *Bothriochloa* toward reduction in size and function of the pedicellate spikelets.

In the two bisexual aneuploids, the B chromosome bearing this gene might be missing. Hybrid 2, however, has the full complement of 40 chromosomes. If the gene in the B genome were missing due to substitution of a D chromosome, the hybrid should look more like *Dichanthium* than it does. The dominant allele might be missing if X-750 were heterozygous for the character. Finally, it may be that there is occasional pairing between B and D chromosomes in X-750 in which preferential pairing is well established, and that the crossover products produced gametes without the gene for suppression of bisexuality in pedicellate spikelets. With present materials and techniques it does not seem possible to decide between the several possibilities.

In the tribe Andropogoneae, the bisexuality of pedicellate spikelets has been used to separate members of the subtribe Saccharinae from those of the subtribes Sorghinae and Andropogoneae. In the case of *D. fecundum*, the character seems to be simply inherited.

Literature cited

- Borgaonkar, D. S. and de Wet, J. M. J. 1961. A cytogenetical study of hybrids between *Dichanthium annulatum* and *D. fecundum*. *Phyton* 15: 137-144.
- Carnahan, H. L. and Hill, H. D. 1961. Cytology and genetics of forage grasses. *Bot. Rev.* 27: 1-162.
- Celazier, R. P. and Harlan, J. R. 1955. An Andropogoneae garden in Oklahoma. *Taxon.* 5: 183-186.
- , de Wet, J. M. J., Borgaonkar, D. S. and Harlan, J. R. 1961. Intergeneric hybrids in Bothriochloinae I. *Bothriochloa intermedia* and *Dichanthium annulatum*. *Cytologia* 26: 170-175.
- Chheda, H. R. 1962. An analysis of hybrids with *Bothriochloa intermedia* (R. Br.) A. Camus. Ph. D. thesis (Unpubl.), Okla. State Univ. pp. 113.
- , de Wet J. M. J. and Harlan, J. R. 1961. Aneuploidy in *Bothriochloa* hybrids. *Caryologia* 14: 205-217.
- de Wet, J. M. J., Borgaonkar, D. S. and Chheda, H. R. 1961. Intergeneric hybrids in Bothriochloinae II. *Bothriochloa* and *Capillipedium*. *Cytologia* 26: 268-273.
- Harlan, J. R., Celazier, R. P., Richardson, W. L., Brooks, M. H. and Mehra, K. L. 1958. Studies on Old World Bluestems II. *Okla. Agri. Expt. Sta. Tech. Bull. T. 72.*
- , de Wet, J. M. J., Richardson, W. L. and Chheda, H. R. 1962. Studies on Old World Bluestems III. *Okla. Agri. Expt. Sta. Tech. Bull. T. 92.*
- Myers, W. M. 1947. Cytology and genetics of forage grasses. *Bot. Rev.* 13: 318-421.
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