

Cytological Study in Hybrids of *Sorghum alnum* (Gramineae)

S. P. SENGUPTA and D. E. WEIBEL

Oklahoma State University, Stillwater

Diploid and tetraploid parental combinations generally result in triploid hybrids, but higher percentages of tetraploids were reported by Endrizzi (1957) and McClure (1965) in hybrids of cultivated diploid sorghum and tetraploid *Sorghum alnum*. Vardi and Zohary (1967) noted this phenomenon in hybrids of wheat and indicated it as diploid introgression into tetraploids. Associated with this phenomenon in sorghum were some cytological behaviors such as: (1) formation of polyploid microsporocytes observed by Damon (1961) in diploid sorghum and by McClure (1965) in hybrids of *S. alnum* and diploid sorghum var. Martin, and (2) asynapsis reported by Stephens and Schertz (1965). The polyploid microsporocytes were classified and named as syncytes after Levan by Price (1956).

This report deals with the chromosome numbers of about 100 hybrids involving cultivated diploid sorghum and tetraploid *Sorghum alnum* and their cytological behavior.

MATERIALS AND METHODS

The materials studied in this report were hybrids of F₁ generation involving diploid cytoplasmic male-sterile varieties of sorghum, namely, Combine Kafir 60, Dwarf Redlan, Martin, Redlan, and Wheatland as female parents and tetraploid *Sorghum alnum* as male parent. The cytological observations were made by studying pollen mother cells stained with acetocarmine.

RESULTS

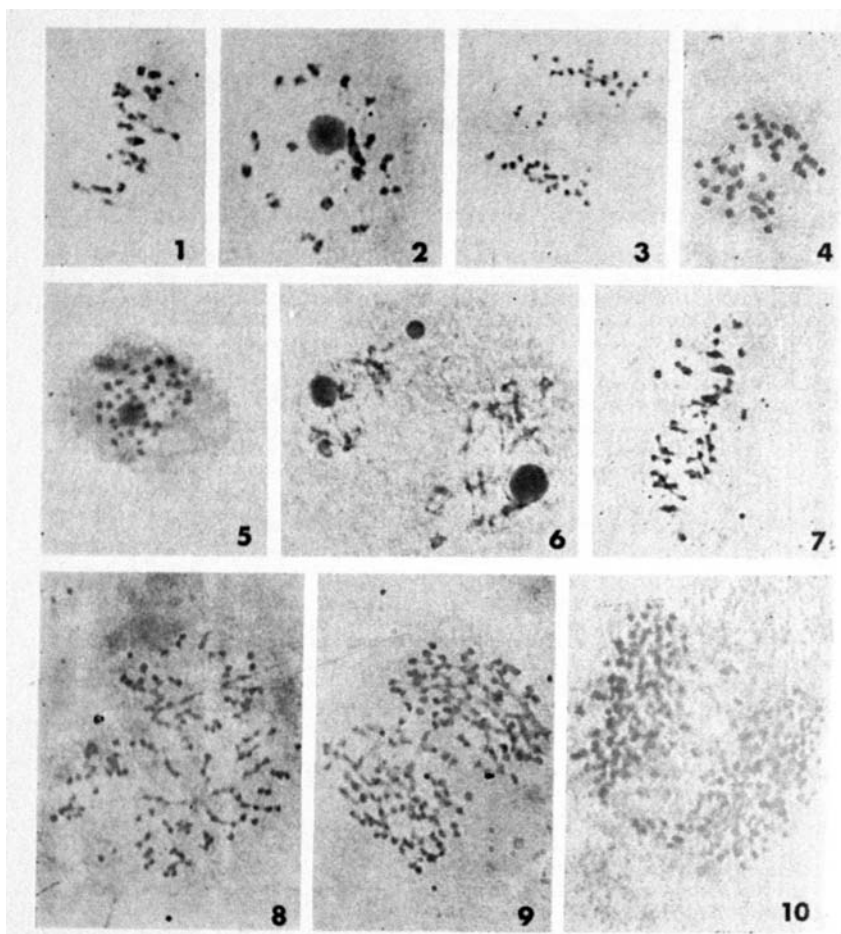
Hybrid combination Martin \times *S. alnum* yielded no triploids, while hybrids in all other combinations yielded triploids and tetraploids (Table I). In triploids chromosome associations showed high incidence of univalents and trivalents averaging up to 3.8 and 3.0 respectively, and occasional quadrivalents (Fig. 1). Bivalent associations averaged up to 10.9. Anaphase I was irregular with laggards. One plant of hybrid combination Dwarf Redlan \times *S. alnum* with regular chromosomes $2n = 30$ showed syncytes with $2n = 60$ chromosomes having an association of 7 univalents, 22 bivalents, and 3 trivalents (Fig. 7). Syncytes with chromosomes $2n = 60$ were also observed in hybrids of Redlan \times *S. alnum* and Wheatland \times *S. alnum*. The triploids were usually male-sterile, the flowering head appearing grassy with no seed-set.

The tetraploids with chromosome $2n = 40$ were found mostly normal with bivalents up to 17.8 and a range from 0 - 20. In some tetraploids one quadrivalent was found to be conspicuously associated with the nucleolus (Fig. 2). Anaphase I was usually regular (Fig. 4), but sometimes irregular with laggards and occasional dividing univalents (Fig. 3) and unequal distribution of chromosomes at the poles. Male fertility was evidenced by grain formation, sometimes with heavy seed-set. In three hybrid combinations few asynaptic cells with 40 univalents were observed (Fig. 5). Two or more nucleoli were occasionally observed in some cells indicating syncyte formation (Fig. 6). One hybrid of Wheatland \times *S. alnum* combination showed syncytes with chromosomes $2n = 80$, 16

TABLE I. CYTOLOGICAL OBSERVATION IN HYBRIDS OF *Sorghum Almum*

Female Parent Common Sorghum Var.	No. of Plants	Chromo- some No. 2n	Av. Chromosome Association* Per Cell Per Plant & Range				Plants with Asynaptic Cells	Syncytes with Chro- mosome 2n
			I	II	III	IV		
Combine Kafir 60	3	30	2.8 0-6	10.9 8-13	0.7 0-4	0.8 0-2	0	0
	14	40	0.6 0-3	16.4 13-20	0.2 0-2	1.5 0-4	0	80
	3	30	2.8 0-8	9.5 5-12	2.1 0-4	0.3 0-2	0	60
Dwarf Redlan	19	40	2.6 0-40	15.3 0-20	0.7 0-4	1.2 0-4	2	0
	20	40	1.6 0-40	17.3 0-20	0.1 0-2	0.9 0-4	2	80, 160 & 200
Redlan	2	30	2.1 0-4	10.4 8-14	1.2 0-2	0.9 0-3	0	60
	17	40	0.5 0-3	16.4 8-20	0.2 0-3	1.6 0-6	0	80
Wheatland	4	30	1.8 0-6	7.8 2-13	3.0 0-8	1.0 0-3	0	60
	16	40	1.2 0-40	17.8 0-20	0.1 0-4	0.7 0-3	1	80, 160 & 300

*I = Univalent, II = Bivalent, III = Trivalent, & IV = Quadrivalent.



LEGEND TO THE FIGURES

- Figure 1. Metaphase with Chromosome $2n = 30$ in hybrid Dwarf Red-lan \times *S. alnum*
- Figure 2. Diakinesis with chromosome $2n = 40$ and one quadrivalent being associated with the nucleolus in hybrid Wheatland \times *S. alnum*.
- Figure 3. Anaphase I with chromosome $2n = 40$ showing laggards and dividing univalents in hybrid Wheatland \times *S. alnum*.
- Figure 4. Anaphase I with 20 chromosomes toward each pole in hybrid Martin \times *S. alnum*.
- Figure 5. Asynaptic cell with 40 univalents in hybrid Martin \times *S. alnum*.
- Figure 6. Syncyte with three nucleoli in hybrid Combine Kafir 60 \times *S. alnum*.

Figure 7. Syncyte with chromosome association 7 univalents, 22 bivalents, 3 trivalents, & 0 quadrivalent at metaphase in hybrid Dwarf Redlan \times *S. alnum*.

Figure 8. Syncyte with chromosomes, $2n = 160$ at metaphase, spreading in stellate fashion in hybrid Wheatland \times *S. alnum*.

Figure 9. Syncyte with chromosome $2n = 160$ at anaphase of the same hybrid as in Figure 8.

Figure 10. Syncyte with chromosome $2n = 200$ approximately at anaphase of hybrid Martin \times *S. alnum*.

(Figs. 8 & 9) and approximately 300 spread over large sporogenous area. In hybrid combination Martin \times *S. alnum* syncytes with chromosomes $2n = 80, 160$ in two plants, and approximately 200 in one plant (Fig. 10) were observed. In one plant of the same hybrid combination semi-asynaptic cells with univalents up to 16 and syncytes with chromosomes $2n = 160$ spreading over large sporogenous areas were observed. Evidently these tetraploid plants were male-sterile. A male-fertile tetraploid of hybrid combination Redlan \times *S. alnum* showed only a few small syncytes.

DISCUSSION AND SUMMARY

Asynaptic cells in three different hybrid combinations ranged from 5 - 10% (Table II). Stephens and Schertz (1965) noted that asynapsis in *Sorghum vulgare* was inherited as a simple recessive and controlled by a single gene giving a 3:1 segregation of fertile to sterile plants. Roy and Jha (1958) studied a plant of *Abelmoschus esculentus*, which was semi-asynaptic and produced large numbers of flowers and fruits. Genes causing complete asynapsis and resulting in polyploid gametes and offspring in several plants were reported (Burnham 1964). These genes might be introduced into any desired variety but would have to be re-

TABLE II. SUMMARY OF OBSERVATIONS

Hybrids	% of Total Plants with					% of Total Tetraploids Male-fertile
	Triploidy	Tetraploidy	Male fertility	Asynaptic cells	Syncytes & Poly- ploidy	
Combine Kafir 60 \times <i>Sorghum alnum</i>	17.6	82.4	47.1	0	5.9- 8X*	50.0
Dwarf Redlan \times <i>Sorghum alnum</i>	13.6	86.4	68.2	9.0	4.5- 6X	73.7
Martin \times <i>Sorghum alnum</i>	0	100.0	55.0	10.0	10.0- 8X, 16X & 20X	55.0
Redlan \times <i>Sorghum alnum</i>	10.5	89.5	52.7	0	10.5- 6X & 8X	52.9
Wheatland \times <i>Sorghum alnum</i>	20.0	80.0	45.0	5.0	10.0- 6X, 8X, 16X, & 30X	50.0

* \times designates sets of haploid chromosomes.

moved from the polyploids produced. If these stocks were crossed with tetraploids, the full-sized seeds would have good chance of being tetraploid. In the present study the plants with asynaptic cells were found to be male-fertile tetraploids, the size of the asynaptic cells being appreciably smaller, and no bigger than the normal meiotic cells. The plant with semi-asynaptic cells and syncytes of larger units (Chromosomes $2n = 160$) was evidently male-sterile.

McClure (1965) observed, in hybrids of Martin \times *S. alnum*, polyploid sporocytes (syncytes), with chromosomes $2n = 50, 80, 100,$ and 120 . He considered that the production of these syncytes might be controlled by the same type of mechanism that resulted in unreduced eggs and also by the breakdown of the cell walls of the microsporocytes. In the present study syncytes were found with chromosomes $2n = 60, 80, 160, 200,$ and 300 in different combination hybrids. This might indicate that these were cases of fusion syncytes, where fusion of cells in varying degrees from smaller to larger units took place by dissolution of the cell walls just prior to meiosis. This was evidenced by the presence of cells with two or more nucleoli and also bigger syncytes with chromosomes of different cell units spread into stellate fashion (Figs. 6 and 8). This was in agreement with the findings of Price (1956), who considered the syncytes in *Saccharum* and *Erianthus* to be cases of fusion syncytes. He suggested that in *Saccharum* and its relatives syncytes might lead to the formation of diploid pollen grains, and they might play a part in the male transmission of $2n$ chromosome numbers.

From the present study the evolutionary implication of the syncytes in the sorghum hybrids appeared to be two fold: (1) the bigger syncytes spreading over larger sporogenous area might degenerate, and (2) the syncytes of smaller units might contribute male diploid pollen to fertilize female gametes of unreduced eggs giving rise to higher polyploids. A certain balance between the asynaptic cells and the syncytes appeared to be necessary to have favorable effect on the fertility of the tetraploids. This was evidenced in the hybrid combination Dwarf Redlan \times *S. alnum*, where as high as 73.7% of the tetraploids were male-fertile, per cent of plants with asynaptic cells being 9 and that with syncytes being only 4.5 (Table II). In a hybrid combination, Wheatland \times *S. alnum*, where the syncytes were larger and covered more sporogenous area, and the incidence of asynapsis was less, male fertility of the tetraploids came down to 50%. An intermediate stage could be noted in hybrid combination Martin \times *S. alnum* where the male fertility of the tetraploids was 55%.

Hybrid combinations of cytoplasmic male-sterile diploid sorghum, namely, Combine Kafir 60, Dwarf Redlan, Martin, Redlan, and Wheatland with tetraploid *Sorghum alnum* constituted 0 - 20% male-sterile triploids and 80 - 100% tetraploids, of which 50 - 73.7% were male-fertile as evidenced by grain formation. Meiosis of microsporogenesis of the male-fertile tetraploids was mostly normal. Percentage of plants with asynaptic cells in 3 hybrid combinations ranged from 5-10, and the asynaptic cells appeared to be smaller in size as compared to normal meiotic cells. Syncytes were found in all combination hybrids with chromosomes $2n = 60, 80, 160,$ approximately 200 and 300, and they were considered to be fusion syncytes. It is suggested that the evolutionary implication of the syncytes in sorghum hybrids could be two-fold: (1) syncytes of larger size might develop nonfunctional pollen of very big size, and (2) syncytes of smaller units might form functional diploid pollen contributing male $2n$ chromosomes and giving rise to higher polyploids. Asynaptic cells appear to counterbalance the size effects of syncytes and favor the formation of male-fertile tetraploids as evidenced in a hybrid combination of Dwarf Redlan \times *Sorghum alnum*.

LITERATURE CITED

- Burnham, C. R. 1964. *Discussion in Cytogenetics*. Burger Publ. Co.
- Damon, E. C. 1961. Studies of the occurrence of multiploid sporocytes in three varieties of cytoplasmic male-sterile and the normal fertile variety, Resistant Wheatland Sorghum. *Phyton* 17 (2):193-203.
- Endrizzi, J. E. 1957. Cytological studies in some species and hybrids in Eu-sorghums. *Bot. Gaz.* 119:1-10.
- McClure, W. J. 1965. Cytological and morphological observations in crosses between diploid and tetraploid sorghum. Unpubl. Ph.D. Thesis, Okla. State Univ.
- Price, Sam. 1956. Cytological studies in *Saccharum* and allied genera 1. Syncytes in certain clones of *Saccharum* and *Erianthus*. *Cytology* 21:21-37.
- Roy, R. P. and R. P. Jha. 1958. A semi-asynaptic of *Abelmoschus esculentus* (L) Moench (= *Hibiscus esculentus* L.) *Cytology* 23:356-361.
- Stephens, J. C. and K. F. Schertz. 1965. Asynapsis and its inheritance in *Sorghum vulgare* Pers. *Crop Sci.* 5:337-339.
- Vardi, A. and D. Zohary. 1967. Introgression in wheat via triploid hybrids. *Heredity* 22:541-560.
-