Cytotaxonomic studies in *Themeda triandra*. I. Chromosome numbers and microsporogenesis

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Metaphase I pairing associations and chiasmata, as well as anaphase I laggard and chromatid bridge data were collected from 53 collections of *Themeda triandra* Forsk. The distribution pattern of the diploids, tetraploids, pentaploids and hexaploids confirms that which was found by previous authors. Diploids occurred mainly in the eastern and southern coastal areas, hexaploids on the highveld and tetraploids in between, but with some overlap. The meiosis analysis suggests that the majority of the polyploids are segmental allopolyploids and that different homeologous genomes occur in this polyploid complex. It is likely that a certain amount of karyotype differentiation must have occurred between different diploid populations of this species.

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Die mikrosporogenese van 53 versamelings van *Themeda triandra* Forsk. is bestudeer. Metafase I-paringsassosiasies en chiasma-analises asook anafase I-sloerder en chromatiedbrug-analises is gedoen van elke versameling. Diploïede, tetraploïede, pentaploïede en heksaploïede is gevind. Die verspreiding van die verskillende poliploïede stem ooreen met die bevindings van vorige outeurs. Diploïede kom langs die suid en oostelike kusgebiede voor, heksaploïede op die hoëveld met die tetraploïede tussen in. Heelwat oorvleueling kom egter voor. Uit die bestudering van meiose is dit duidelik dat die meerderheid van die poliploïede segmentele-allopoliploïede is. Dit dui op kariotipe-differensiasie tussen verskillende diploïede bevolkings.

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Introduction

Embryo sac development studies on *Themeda triandra* Forsk. have indicated that this important South African grass probably forms an agamic complex. Brown and Emery (1957, 1958) reported that a tetraploid from South Africa and an octoploid from India were both aposporic apomicts according to the terminology proposed by Stebbins (1950). These findings have been supported by myself (Liebenberg 1961; Liebenberg & Pienaar 1962), as well as by Birari (1980) who studied 4x, 8x, 9x and 11x *T. triandra* collections from India. However, a diploid form of *T. triandra* has been reported with normal sexual embryo sac development of the *Polygonum* type (Liebenberg 1961). Studies on other *Themeda* species from Asia and Australia have shown the same pattern (Woodland 1964; Faruqi *et al.* 1975; Birari 1980).

Cytogenetic studies by Gluckmann (1951), de Wet (1960), Tateoka (1965), Gould & Soderstrom (1974), Mehra and Sharma (1975), Birari (1981) and Birari (1983) have shown that *T. triandra* is a polyploid complex as is characteristic of most agamic complexes. As far as southern Africa is concerned these results show that polyploid levels of 2n = 2x, 3x, 4x, 5x and 6x occur, including a number of aneuploids. Diploids seem to occur mostly along the eastern and southern coastal areas, the hexaploids mainly on the highveld whereas the tetraploids tend to occupy intermediate areas.

Only Gluckmann (1951) gives some information on the meisosis of South African *T. triandra*. Her studies on meiosis were from plants from a limited number of localities and her results incomplete. Her general conclusions were that the even-polyploids showed a high level of bivalent pairing. Somewhat more information is available on the meiosis of *T. triandra* collections as well as other *Themeda* species from India and Australia (Brown & Emery 1957; Hayman 1960; Raman *et al.* 1959; Sisodia 1971; Birari 1981).

The present study was undertaken in an attempt to gather more information on the microsporogenesis and embryo sac development of *T. triandra* in southern Africa. Hopefully such information will eventually lead to a better understanding of the origin of this agamic complex. The present report deals with chromosome numbers and microsporogenesis (meiosis) of 53 collections while future reports will deal with population studies and the embryo sac development.

Materials and Methods

Fifty-three plants collected by Williams from various localities in South Africa were studied (Table 1). Herbarium specimens of these plants have been placed in the National Herbarium at Pretoria. For cytogenetic studies young inflorenscences were

Table 1 Origin of 53 collections of *Themeda triandra* and summary of metaphase I analysis

Collection No. *Williams*	Locality	2n	$\bar{x}$ metaphase I Xta & stand dev.	Xta per chromosome association (a)	$\bar{x}$ metaphase I pairing associations				$\bar{x}$ metaphase I laggards	% of chromosomes bound as bivalents, etc.			
					II	I	III	IV		II	I	III	IV
2	29 km W. of Worcester towards Du Toitskloof	20	15,9 ± 0,67	1,59	10,00				0,00	100,0			
7A	10 km N. of Malmesbury	20	14,1 ± 0,42	1,41	10,00				0,00	100,0			
11	Cable station Table Mountain, Cape Town	20	14,2 ± 0,44	1,42	10,00				0,00	100,0			
13	20 km SW Somerset West	20	14,0 ± 0,44	1,40	10,00				0,00	100,0			
18	25 km W. of Mossel Bay towards Riversdale	20+4B	14,4 ± 0,47	1,44	10,00				0,00	100,0			
19	Outeniqua Research Station, George	20	14,4 ± 0,54	1,45	9,98	0,08			0,08	99,6	0,4		
24	6 km W. of Bedford towards Adelaide	20+6B	14,4 ± 0,42	1,44	10,00				0,00	100,0			
28A	40 km S. of Grahamstown on farm 'Vendo'	20	14,1 ± 0,59	1,41	10,00				0,16	100,0			
29	Bathurst Res. Station	20	14,3 ± 0,43	1,44	9,96	0,08			0,00	99,6	0,4		
31	5 km NE of Peddie towards Berlin	20	13,6 ± 0,37	1,36	10,00				0,08	100,0			
60	Foothills of Mont-aux-Sources	20+4B	13,9 ± 0,52	1,39	10,00				0,00	100,0			
65	Grootfontein Res. Station, Middelburg (Cape)	20	14,1 ± 0,39	1,42	9,96	0,08			0,00	99,6	0,4		
76A	Welgevallen Res. Station, Stellenbosch	20	15,2 ± 0,41	1,52	10,00				0,00	100,0			
91	Vaalharts Res. Station, Christiana	20	14,7 ± 0,49	1,47	10,00				0,00	100,0			
246	Richards Bay	20	13,9 ± 0,59	1,39	10,00				0,00	100,0			
4	88 km SE of Worcester towards Bonnievale	21	12,7 ± 0,53	1,27	9,88	0,08	0,12		0,92	94,1	4,2	1,7	
10	27 km S. of Porterville towards Gouda	21	13,2 ± 0,50	1,32	10,00	1,00			0,68	95,2	4,8		
70	Swaershoek Pass, Cradock	21	12,4 ± 0,41	1,24	9,92	0,92	0,08		0,04	94,5	4,4	1,1	
63/67	Between Crecy and Naboomspruit	23	13,8 ± 0,47	1,28	10,48	1,00	0,28	(b)	1,08	91,0	4,3	3,6	(b)
32	46 km E. of East London towards Peddie	40	24,8 ± 0,73	1,30	18,20	0,90	0,80	0,12	0,40	90,8	2,3	5,7	1,2
34	3 km W. of Barkly East towards Queenstown	40	24,8 ± 0,65	1,26	18,6	0,32	0,32	0,36	0,28	92,8	0,8	2,4	4,0
37	5 km W. of Lady Grey towards Jamestown	40	27,5 ± 0,70	1,39	16,5	0,28	0,28	1,50	0,24	82,4	0,7	2,1	14,8
38	Ukulinga Res. Station, Pietermaritzburg	40	27,1 ± 0,68	1,36	18,7	0,08	0,08	0,56	0,12	93,6	0,2	0,6	5,6
40	Zululand (?)	40	27,1 ± 0,90	1,39	17,9	0,44	0,20	0,80	0,52	89,4	1,1	1,5	8,0
46A	Escourt Res. Station	40	27,0 ± 0,88	1,37	17,2	0,36	0,28	1,08	0,48	86,2	0,9	2,1	10,8
54	Cathedral Peak	40	24,9 ± 0,84	1,27	18,2	0,32	0,24	0,64	0,40	91,0	0,8	1,8	6,4
64	Grootfontein Res. Station, Middelburg (Cape)	40	26,9 ± 0,77	1,36	16,0	0,24	0,24	1,40	0,44	83,6	0,6	1,8	14,0
72	16 km NE of Graaff-Reinet	40	25,0 ± 0,75	1,30	17,1	0,84	0,16	0,68	0,60	85,4	2,1	5,7	6,8
73	21 km NE of Colesberg towards Philippolis	40	28,1 ± 1,00	1,43	16,5	0,40	0,40	1,36	0,32	82,4	1,0	3,0	13,6
75	Penhoek Pass between Aliwal North and Queenstown	40	24,0 ± 0,82	1,22	19,5	0,40	0,32	0,08	0,16	95,8	1,0	2,4	0,8
86	Glen Res. Station, Bloemfontein	40	25,8 ± 0,75	1,31	16,6	0,36	0,36	0,36	0,32	82,8	0,9	2,7	13,6
88	11 km S. of Petrusberg, Fauresmith	40	25,3 ± 0,96	1,26	17,4	0,96	0,80	0,40	1,00	87,2	2,5	6,3	4,0
90	Koopmansfontein Res. Station, Barkly West	40	23,8 ± 0,64	1,19	18,8	0,36	0,28	0,28	0,56	94,2	0,9	2,1	2,8
97	8 km NE of Weenen	40	24,4 ± 0,79	1,22	18,9	0,24	0,16	0,36	0,24	94,6	0,6	1,2	3,6
98	13 km NE of Weenen	40	25,8 ± 0,79	1,29	16,5	0,60	0,60	1,16	0,72	82,4	1,5	4,5	11,6
99GR	Magut, northern Natal	40	22,7 ± 0,75	1,13	18,8	1,12	0,40	0,04	0,72	93,8	2,8	3,0	0,4
103	8 km S. of Nelspruit towards Kaapsehoop	40	24,6 ± 0,74	1,23	17,9	1,24	0,52	0,32	1,52	89,8	3,1	3,9	3,2
261	Between Piet Retief and Pongola	40	26,3 ± 0,70	1,31	18,7	0,28	0,28	0,36	0,32	93,6	0,7	2,1	3,6
65/9	Mac-Mac Falls, Sabie	40+4B	25,1 ± 0,69	1,25	18,9	0,68	0,52	–	0,68	94,4	1,7	3,9	–
21	St John's-drif, 22 km E. of Dohne	42	23,7 ± 0,49	1,12	20,4	0,44	0,20	0,04	0,28	97,1	1,1	1,4	0,4
A131	Condover, Zululand	42	24,8 ± 0,85	1,18	20,0	1,52	0,08	0,04	1,24	95,4	3,6	0,6	0,4
78	Athole Res. Station, Ermelo	50	27,5 ± 0,69	1,37	17,0	6,28	2,76	0,36	5,36	68,0	12,6	16,6	2,9
84	3 km NW of Zastron	50	27,0 ± 0,96	1,38	16,5	7,64	2,68	0,32	6,32	66,1	15,3	16,1	2,6
99BL	Magut, northern Natal	50	26,3 ± 1,30	1,34	17,8	8,80	1,84	0,04	8,56	71,0	17,6	11,0	0,3
82	Lesotho border road to Mafekeng	59	33,7 ± 0,72	1,16	27,4	1,36	0,84	0,08	1,36	92,9	2,3	4,3	0,5
45	13 km E. of Howick	60	36,6 ± 1,10	1,22	20,0	4,00	3,40	1,48(b)	4,20	66,8	6,7	16,9	9,4(b)
77	Kosmos, Hartebeespoort Dam	60	36,9 ± 0,80	1,23	28,2	1,20	0,56	0,16	1,72	94,1	2,0	2,8	1,1

Table 1 (Continued)

Collection No. *Williams*	Locality	2n	$\bar{x}$ metaphase I Xta & stand dev.	Xta per chromosome association (a)	$\bar{x}$ metaphase I pairing associations				$\bar{x}$ metaphase I laggards	% of chromosomes bound as bivalents, etc.			
					II	I	III	IV		II	I	III	IV
80	42 km W. of Rustenburg towards Swartruggens	60	35,1 ± 0,75	1,17	29,2	0,52	0,28	0,08	0,76	97,2	0,8	1,4	0,5
81	58 km SE of Bloemfontein towards Reddersburg	60	35,0 ± 0,71	1,16	27,3	1,36	0,96	0,28	2,10	91,1	2,3	4,8	1,9
85	3 km SW of Aliwal North	60	34,3 ± 0,63	1,14	27,6	2,08	0,88	–	1,68	92,1	3,5	4,4	–
96	Valsch River Dam, Kroonstad	60	35,0 ± 0,77	1,16	28,7	1,04	0,48	0,04	1,40	95,6	1,7	2,4	0,3
63/71	8 km NW of Naboomspruit	60	35,8 ± 0,98	1,19	27,1	1,36	1,12	0,28	1,36	90,3	2,3	5,6	1,9
Pta	Rietondale Res. Station, Pretoria	60	37,1 ± 0,78	1,27	28,4	0,56	0,32	0,40	0,20	94,8	0,9	1,6	2,7

(a) Calculated as $\bar{x}$ Xta ÷ ($\bar{x}$ number of II + III + IV at metaphase I)
(b) These two collections also had one meiocyte each having a pentavalent association

fixed in 6 parts methanol : 3 parts chloroform : 2 parts propionic acid and squashed in 1% propionic carmine (Pienaar 1955). Although meiosis was studied in its entirety, only metaphase I : pairing associations and chiasma analysis; and anaphase I laggards and chromatid and chromosome bridges are described here. Twenty-five cells of each stage were studied.

Results

The results of the metaphase I and anaphase I analysis are summarized in Tables 1 & 2. Metaphase I pairing associations of the tetra- and hexaploids are given in more detail in Tables 3 & 4.

Of the 53 collections studied, 15 (28,3%) were diploids (2n = 20 = 2x); 20 (37,7%) were tetraploids (2n = 40 = 4x); 3 (5,7%) were pentaploids (2n = 50 = 5x); 8 (15,1%) were hexaploids (2n = 60 = 6x); the remaining 7 were aneuploids: 3 (5,7%) trisomics (2n = 21 = 2x + 1); 1 (1,9%) had 2n = 23 = 2x + ?; 2 (3,8%) had 2n = 42 = 4x + 2; 1 (1,9%) had 2n = 59 = 6x − 1. Furthermore, 4 collections (*Williams, 18, 24, 60* and *65/9*) also possessed B (supernumary) chromosomes.

As can be expected, the meiosis in the diploids is normal (10_{II} at metaphase I and regular segregation at anaphase I). The only abnormality encountered in the diploids is the rare occurence at metaphase I of 2 univalents or at anaphase I of 2 laggards. This was observed in 5 of the 15 diploids (Tables 1 & 2). At metaphase I three collections (*Williams 19, 29,* and *65*) each had one meiocyte (4%) with $9_{II} + 2_{I}$ (Table 1). At anaphase I three collections (*Williams 19, 28A* and *31*) each had one meiocyte (4%) possessing 2 laggards. Only *Williams 19* had both a metaphase I and an anaphase I with this abnormality. From these results it would seem that the asynapsis of one chomosome pair is a phenomenon that occurs sporadically in most if not all diploids. In all, 750 metaphase I and anaphase I meiocytes of the 15 diploids were studied. Of these, 7 (0,93%) exhibited asynapsis of one chromosome pair.

Most tetraploid collections possess a surprisingly normal meiosis although some variation occurs as regards pairing associations (Tables 1, 2 & 3). The percentage of chromosomes paired as bivalents exceeds 82% in all tetraploids and is as high as 94% in some (e.g. *Williams 90, 97,* and *65/9*). The proportion of unbound chromosome univalents is low (from 0,2% in *Williams 38* to 3,1% in *Williams 103*).

The variation that exists between the meiosis of the 20 different tetraploids can best be seen in a comparison of their complete pairing associations (Table 3). From these results it can be seen that some collections exhibit a high proportion of meiocytes with quadrivalents and fairly low proportions of trivalents (e.g. *Williams 37, 38, 40, 46A* and *64, 73, 86, 98* amongst others), whereas other collections show the exact opposite (e.g. *Williams 32, 72, 75, 88, 90, 99GR* and 65/9).

Two of the tetraploids (*Williams 32* and *72*) also showed heterozygosity for a paracentric inversion (Table 2).

As would be expected, the meiosis in the pentaploids proved to be very irregular (Tables 1 & 2). Low percentages of the chromosomes being paired as bivalents and there were relatively many univalents and trivalents. Quadrivalents occur at about the same frequencies as they do in the tetraploids while no multivalents of a higher order than four could be identified with any degree of certainty. The pairing associations are extremely variable in the pentaploids.

Meiosis in the hexaploids appears to be more regular than in the tetraploids (Tables 1, 2 & 4). The one exception, *Williams 45*, has an even more irregular meiosis than in the pentaploids. The other hexaploids all have meiocytes with more than 90% of the chromosomes forming bivalents. In *Williams 80*, collected at Rustenburg, Transvaal, bivalent formation was as high as 97,2%. The hexaploids studied have different patterns of chromosome association and this is summarized in Table 4.

The 3 trisomics (*Williams 10* and *70*) are all probably primary trisomics. The only pairing associations that were encountered being $10_{II} + 1_{I}$ and $9_{II} + 1_{III}$. The majority of metaphase I's possessed the first type (*Williams 4* = 88% of the meiocytes, *Williams 10* = 100% and *Williams 70* = 92%).

The aneuploid *Williams 63/77* (2n = 23) is much more difficult to classify. One meiocyte showed a possible pentavalent at metaphase I. If this analysis is correct, the plant must be a pentasomic (2n = 23 = 2x + 3), but it could also be tetrasomic for one chromosome and trisomic for another, (2n = 23 = 2x + 2 + 1); the pairing associations that were found being $11_{II} + 1_{I}$ (52%); $10_{II} + 3_{I}$ (16%); $10_{II} + 1_{III}$ (28%) and $9_{II} + 1_{V}$ (?) (4%).

The two plants which have 2n = 42 (*Williams 21* and *A131*) show slightly different pairing associations. The latter has a

Table 2 Analysis of unbound chromosomes at metaphase I and anaphase I

Collection No. *Williams*	2n	$\bar{x}$ monovalents at metaphase I	$\bar{x}$ laggards at anaphase I	% of meiocytes with different numbers of unbound chromosomes per meiocyte at metaphase I and anaphase I metaphase I monovalents / anaphase laggards 0	1	2	3	4	5	6	7	8	9	10	11	12	17
2	20	0,00	0,00	100/100													
7A	20	0,00	0,00	100/100													
11	20	0,00	0,00	100/100													
13	20	0,00	0,00	100/100													
18	20+4B	0,00	0,00	100/100													
19	20	0,08	0,08	96/96		4/4											
24	20+6B	0,00	0,00	100/100													
28A	20	0,00	0,16	100/92		0/8											
29	20	0,08	0,00	96/100		4/0											
31	20	0,00	0,08	100/96		0/4											
60	20+4B	0,00	0,00	100/100													
65	20	0,08	0,00	96/100		4/0											
76A	20	0,00	0,00	100/100													
91	20	0,00	0,00	100/100													
246	20	0,00	0,00	100/100													
4	21	0,88	0,92	12/8	88/92												
10	21	1,00	0,68	0/32	100/68												
70	21	0,92	0,04	8/96	92/4												
63/67	23	1,00	1,08	32/32	52/48		16/20										
32	40	0,90	0,40	48/80	24/8	20/4	4/8	4/0	(+ 20% chromatid bridges)								
34	40	0,32	0,28	68/76	32/20	0/4											
37	40	0,28	0,24	72/80	28/16	0/4											
38	40	0,08	0,12	92/92	8/4	0/4											
40	40	0,44	0,52	80/76	8/16	8/0	0/4		4/0	0/4							
46A	40	0,36	0,48	72/64	20/24	8/12											
54	40	0,32	0,40	72/72	24/24	4/0		0/4									
64	40	0,24	0,44	76/68	24/24	0/4	0/4										
72	40	0,84	0,60	32/60	52/24	16/12	0/4	(+ 40% chromatid bridges)									
73	40	0,40	0,32	64/72	32/24	4/4											
75	40	0,40	0,16	68/88	24./8	8/4											
86	40	0,36	0,32	64/76	36./16	0/8											
88	40	0,96	1,00	44/52	32/16	16/20	0/8	4/0	4/4								
90	40	0,36	0,56	68/64	28/20	4/12	0/4										
97	40	0,24	0,24	84/80	8/16	8/4											
98	40	0,60	0,72	52/60	36/16	12/20		0/4									
99GR	40	1,12	0,72	40/32	32/28	20/8	0/4	4/4		4/0							
103	40	1,24	1,52	32/28	28/24	28/28	8/12	4/4	0/4								
261	40	0,28	0,32	72/76	28/16	0/8											
65/9	40+4B	0,68	0,68	56/72	28/8	8/4	8/12	0/4									
21	42	0,44	0,28	76/76	8/20	12/4	4/0	–									
A131	42	1,52	1,24	40/44	8/16	32/24	0/4	20/12									
78	50	6,28	5,36			4/0	0/4	8/28	20/20	24/36	20/4	12/4	8/4				
84	50	7,64	6,32				0/8	4/16	4/8	8/16	28/16	24/28	28/8	4/0			
99BL	50	8,80	8,56					0/4	0/16	4/0	4/4	44/20	20/24	16/24	8/4	4/0	0/4
82	59	1,36	1,38	16/18	44/56	28/16	12/4	0/4	0/4								
45	60	4,00	4,20		4/12	20/12	12/8	24/16	24/28	12/16	0/4	0/4	4/0	(+ 36% chromatid bridges)			
77	60	1,20	1,72	32/16	36/32	20/28	4/12	8/4	0/4								
80	60	0,52	0,76	68/68	20/16	8/8	0/4	4/0				0/4					

Table 2 (Continued)

Collection No. *Williams*	2n	$\bar{x}$ monovalents at metaphase I	$\bar{x}$ laggards at anaphase I	% of meiocytes with different numbers of unbound chromosomes per meiocyte at metaphase I and anaphase I (metaphase I monovalents / anaphase laggards)													
				0	1	2	3	4	5	6	7	8	9	10	11	12	17
81	60	1,36	2,10	28/20	28/24	28/24	12/12	4/8	0/4	0/4	0/4						
85	60	2,08	1,68	20/28	16/24	20/24	28/20	12/8	4/0								
96	60	1,04	1,40	44/36	24/8	24/44	0/4	8/8									
63/71	60	1,36	1,36	20/36	36/24	36/28	4/0	4/4	0/8								
Pta.	60	0,56	0,20	64/84	20/12	12/4	4/0										

Table 3 The frequencies of the various metaphase I pairing associations of the tetraploids

Metaphase I pairing associations	Percentage of metaphase I microsporemothercells of each tetraploid collection with different pairing associations																				% collections in which found
	32	34	37	38	40	46A	54	64	72	73	75	86	88	90	97	98	99GR	103	261	65/9	
20_{II}	40	40	12	56	44	16	40	16	16	12	64	8	32	52	60	4	36	20	56	56	100
$19_{II} + 2_{I}$											4			4	4		20	20			25
$18_{II} + 1_{I} + 1_{III}$	24	24	4	4		8	12		32	12	24		20	24	4	20	32	16	16	28	85
$18_{II} + 1_{IV}$	8	24	20	28	20	24	20	24	4	20	4	40	12	8	20	32	4	12	16		95
$18_{II} + 4_{I}$																		4			5
$17_{II} + 3_{I} + 1_{III}$																				8	5
$17_{II} + 2_{I} + 1_{IV}$	4				4	4	4		4												25
$17_{II} + 6_{I}$ •																	4				5
$16_{II} + 2_{I} + 2_{III}$	16				4	4							8		4			8		8	35
$16_{II} + 1_{I} + 1_{III} + 1_{IV}$		8	20		4	12	12	12	16	8		20	8	4		4		12	8		70
$16_{II} + 2_{IV}$		4	24	4	8	28	8	28	8	20		8		4	4	12					65
$15_{II} + 4_{I} + 2_{III}$	4																4				10
$15_{II} + 3_{I} + 1_{III} + 1_{IV}$																		8			5
$14_{II} + 3_{I} + 3_{III}$	4																				5
$14_{II} + 3_{IV}$			16	4	8	4	4	8		8		8				4					45
$14_{II} + 2_{I} + 2_{III} + 1_{IV}$									8	4	4		8			8					25
$14_{II} + 1_{I} + 1_{III} + 2_{IV}$				4	4			12	4	12		12	4		4	8					45
$14_{II} + 5_{I} + 1_{III} + 1_{IV}$					4								4								10
$12_{II} + 4_{I} + 4_{III}$													4								5
$12_{II} + 2_{I} + 2_{III} + 2_{IV}$									4							4					10
$12_{II} + 1_{I} + 1_{III} + 3_{IV}$			4					4								4			4		20
$12_{II} + 4_{IV}$										4											5
Number of different types of associations	7	5	7	6	9	8	7	6	9	9	5	7	9	6	7	10	6	8	5	4	

higher percentage of unbound chromosomes (3,6%) than the former (1,1%). In both however, the extra two chromosomes are homologous as shown by the fact that in both the most common type of pairing association is 21_{II} (*Williams A131* = 40%; *Williams 21* = 72%). Both may therefore be described as tetrasomic tetraploids (2n = 42 = 4x + 2). Meiosis in the monosomic hexaploid, *Williams 82* (2n = 59 = 6x − 1), corresponds very well with that of the majority of the hexaploids — except that it lacks one chromosome.

In general the anaphase I laggard analysis correlates very well with the metaphase I univalents (Table 2). A highly significant correlation was calculated (r = 0,8380). From this one can deduce that although only 25 meiocytes per stage were analysed, the results do show a high repeatability. However there are some disturbing discrepancies. The most conspicuous example of this being two of the trisomics (*Williams 10* and *70*) where the metaphase I and anaphase I results were for some or other reason in direct contradiction with each other.

The laggards show chromatid segregation at both anaphase I and II, a phenomenon that is not unknown in grasses and

Table 4 The frequencies of the various metaphase I pairing associations of the hexaploids

Metaphase I pairing associations	Percentage of metaphase I microsporemother-cells of each collection with different pairing associations							% collections in which found
	77	80	81	85	96	63/71	Pta	
30_{II}	20	64	20	20	44	16	32	100,0
$29_{II} + 2_{I}$	12	4		12	16	4	8	85,7
$28_{II} + 1_{I} + 1_{III}$	32	16	16	16	20	24	20	100,0
$28_{II} + 1_{IV}$	12	4	8			4	24	71,4
$28_{II} + 4_{I}$	8	4	4		4			57,1
$27_{II} + 3_{I} + 1_{III}$	4		8	28			4	57,1
$27_{II} + 2_{I} + 1_{IV}$			4					14,3
$26_{II} + 2_{I} + 2_{III}$	8	4	20	8	8	24	8	100,0
$26_{II} + 1_{I} + 1_{III} + 1_{IV}$	4	4	12		4	12		71,4
$26_{II} + 2_{IV}$							8	14,3
$26_{II} + 5_{I} + 1_{III}$				4				14,3
$25_{II} + 4_{I} + 2_{III}$				12	4	4		42,9
$25_{II} + 3_{I} + 1_{III} + 1_{IV}$						4		14,3
$24_{II} + 3_{I} + 3_{III}$			4					14,3
$24_{II} + 2_{I} + 2_{III} + 1_{IV}$			4			8		28,6
Number of different types of associations	8	7	10	7	7	9	7	

which Gluckmann (1951) also noted in *Themeda*.

Apart from the two tetraploids already mentioned (*Williams 32 & 72*), the very abnormal hexaploid (*Williams 45*) was the only other plant to exhibit paracentric inversion heterozygosity.

The average metaphase I chiasma analysis varied between 13,6 (*Williams 31*) and 15,9 (*Williams 2*) for the diploids, 22,7 (*Williams 99GR*) and 28,7 (*Williams 73*) for the tetraploids, 26,3 (*Williams 99BL*) and 27,5 (*Williams 78*) for the three pentaploids and between 34,4 (*Williams 85*) and 37,1 (plant Pta) for the hexaploids (Table 1). The mean number of chiasmata per average chromosome association (see Table 1 for calculation) ranges between 1,36 and 1,59 for diploids; 1,2 and 1,43 for tetraploids and 1,19 (*Williams 80*) and 1,41 (*Williams 45*) for hexaploids. In the hexaploids it is interesting to note that the plants having the lowest and highest average number of chiasmata per meiocyte do not necessarily have the highest and lowest average number of chiasmata per average chromosome association. This is due to the interaction between low number of unpaired chromosomes and higher percentage of chromosomes bound as quadrivalents and vice versa. Generally speaking, it can be seen that the number of chiasmata per average chromosome association tends to be highest in the diploids and lowest in the hexaploids. A certain amount of overlap does, however, occur. Aneuploids tend to have less chiasmata than their corresponding euploids. The chiasma frequencies found in this study are generally lower than that reported for *T. tremula* and *T. anathera* (Sisodia 1971).

The behaviour of the B-chromosomes was studied in some detail in *Williams 65/9* (2n = 40 + 4B). Most, if not all, meiocytes had four supernumary chromosomes. In the 25 meicotyes studied the B-chromosomes occurred as 4_{I} in 24% of the cells; as 2_{II} in 20%; as $1_{II} + 2_{I}$ in 44% and as $1_{III} + 1_{I}$ in 12% of the cells. The four B-chromosomes in this plant thus appear to be homologous. The other plants with B-chromosomes (*Williams 18, 24* and *60*) had similar pairing associations. Birari (1981) also reports synapsis between B-chromosomes of *Themeda* species in India.

Discussion

Although this study is the most extensive of its kind on the cytogenetics (particularly microsporogenesis) of *Themeda triandra*, it is clear that it does not include enough collections to obtain a reliable representation of the true cytogenetic situation in this agamic complex. Clearly cytogenetic differences exist between different collections of the same polyploid level, especially in the tetraploids. These differences, however, show no clear geographical or other pattern. This could either be due to a too small sample size, or it might reflect a lack of order that does in fact exist and which would not be unexpected in an agamic complex (Stebbins 1950; Grant 1971).

The distribution of the different polyploids generally follows the pattern described by Gluckmann (1951) and De Wet (1960). No triploids were found. This was surprising since De Wet (1960) reported triploids occurring in all the veld types of South Africa. It must be stressed that De Wet based his study largely on stomatal length measurements. Gluckmann (1951) encountered triploids only in Griqualand West. None of the authors reporting on chromosome numbers of Indian *Themeda* species found triploids. Hayman (1960) reported two triploids from only one area in Australia. Further information on the status of triploids in *T. triandra* is required, particularly in the light of the theories put forward by De Wet (1980) on the role played by fertilization by non-reduced gametes in the evolution of polyploid complexes. There is a tendency for the polyploids to increase in frequency with elevation away from

the coast, but there are areas such as the northern Transvaal where the distribution pattern of the diploids and polyploids does not follow this pattern.

The meiosis of the diploids is essentially regular. No clear indications of any structural hybridity could be found. The very low frequency of one pair of chromosomes that are unpaired at metaphase I, found in five of the diploids, tends to suggest that this is a normal phenomenon of all diploids. Whether this reflects some structural heterozygosity which is not expressed in any other way, is difficult to say. Due to the chromatid segregation that occurs at both anaphase I and II, the two monovalents could result in aneuploid gametes ($n = 11 = x + 1$). This could explain the reasonably high frequency of trisomics found in this study as well as by Gluckmann (1951).

Most, if not all of the polyploids, are probably segmental allopolyploids (defined here as a hybrid polyploid with homeologous genomes between which allosyndetic pairing occurs, whereas an allopolyploid would have no allosyndetic pairing between its homeologues). This implies that structural differences occur between the constituent genomes. This in no way presumes that the parents of the polyploid hybrids belong to different species, but that they may be hybrids between differently adapted populations belonging to the same species. It is generally accepted that such hybrids play an important role in the evolution of polyploid complexes (Stebbins 1985). However, due to the structural differences that apparently exist between at least some of the genomes of *T. triandra*, the genomes are homeologous and such polyploids cannot be called autopolyploids. The segmental allopolyploid nature of the *T. triandra* polyploids is supported by the fact that their meiosis indicates that some structural hybridity exists between the chromosomes. Three of the collections (*Williams 32, 72* and *45*) are heterozygous for a paracentric inversion. Although the proof is somewhat circumstantial, heterozygous translocations seem to be common. This can be deduced if a closer inspection of the meiosis of the tetraploids in particular is made. The data (Table 3), suggests that the tetraploids fall into three major groups on the basis of their pairing associations. The first comprises plants exhibiting a high percentage of meiocytes with 20_{II} and a high percentage of meiocytes with an equal number of univalents and trivalents. Quadrivalents occur at a low frequency. Belonging to this group would be collections *32, 75, 88, 90, 99GR, 65/9, 103* and *261*. The aneuploid *Williams 21* probably also belongs to this group.

The second group comprises plants with a relatively low percentage of meiocytes with 20_{II} but a high percentage of cells containing quadrivalents. *Williams 37, 46A, 64, 73, 86* and *98* belong to this group. Group three also shows a high proportion of meiocytes with quadrivalents but differs from the second group in having a high proportion of cells with 20_{II} (*Williams 34, 38, 40, 54* and *97*).

Williams 72 does not fit well into any of these three groups. The existence of tetraploids with differing patterns of pairing associations makes it highly unlikely that all the plants are autopolyploids although some of them might be. A much more uniform pairing pattern would be expected in that case. The differences that do occur would seem to indicate that those plants which are more prone to form quadrivalents are probably heterozygous for reciprocal translocations, whereas the others are not. This is supported by the fact that in the trisomic plants as well as *Williams 63/67* ($2n = 23$) where a chromosome occurs in a polysomic state, these multiple chromosomes very seldom form multivalents. From these arguments it would seem that the different tetraploid groups are made up of slightly different genomes.

The distribution of the plants belonging to the abovementioned three groups shows some interesting, but not always sharp, patterns. Plants belonging to the first group occur in two widely separated areas, one in the eastern Transvaal – northern Natal, and the other in a line running from Barkly West to East London.

The second group occurs primarily in the southern Orange Free State and the neighbouring areas of the Cape Province. Two collections of this group came from the Natal Midlands (*Williams 46A* and *98*).

The third group occurs east of the Drakensberg in Natal, south to the eastern Cape Province. It is not yet possible to determine the possible origin of some of the diploid progenitors of the tetraploids without any experimental cytotaxonomic data. Furthermore, as mentioned previously, it is not impossible that some of the tetraploids might be autotetraploids. Since it is not known at this stage what the expected pairing associations of an autotetraploid in *Themeda triandra* would be, it could very well be that one of the abovementioned groups are autotetraploids. Unfortunately, attempts to produce an autotetraploid by chromosome doubling have not as yet succeeded.

It was more difficult to group the nine hexaploids than the tetraploids (including the aneuploid *Williams 82*). All hexaploids, with the exception of *Williams 45*, show a high frequency of bivalent pairing while quadrivalent formation is comparatively low. *Williams 45* has a completely different type of meiosis to that of any other hexaploid or even tetraploid. The meiosis is much more abnormal with much higher percentages of chromosomes being unbound or bound as multivalents. This plant is the only hexaploid possessing inversion bridges and fragments which indicates that it is probably a segmental allohexaploid.

From the results to date, it is reasonably certain that the genome constitution of the different segmental allopolyploids and possible autopolyploids within a particular polyploid level, varies from area to area. It would seem that some karyotype evolution has occurred in the divergence of the different diploid biotypes and that the hybrid complex is therefore made up of various closely related homeologous and homologous genomes (e.g. A_1, A_2, A_3, etc.).

The studies of the meiosis of *Themeda triandra* as well as other *Themeda* species from India, substantiates this conclusion that the polyploids are segmental allopolyploids and therefore of hybrid origin. Sisodia (1971) reports chromatid bridges and fragments as well as heteromorphic bivalents, both pointing to structural hybridity. Birari (1980, 1981) also concludes that the *Themeda* polyploids that he studied are segmental allopolyploids.

In conclusion, it can be said that much can be gained from studying the meiosis of *Themeda triandra* in an attempt to solve the evolutionary development of this polyploid complex. Because these results are variable, it is clear that similar studies of larger samples from the whole distribution range of the species as well as more detailed studies of local populations are necessary before cytotaxonomic deductions of any value can be made. Forthcoming reports in this series will report on further studies in this respect and further discussions on the possible evolution of *T. triandra* would then be more appropriate.

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