Chromosome Pairing in Interspecific Hybrids of *Tagetes patula* and *T. erecta*

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Tagetes is a genus of the subfamily Heleniae of the ligulate Compositae. Diploid *T. erecta* ($2n=24$) and tetraploid *T. patula* ($2n=48$) have been grown as ornamental plants. Eyster (1941) suggested that *T. patula* was derived from chromosome doubling of *T. erecta*, but Bolz (1961) and Towner (1961, 1962) suggested that *T. patula* was an amphidiploid from *T. erecta* and *T. tenuifolia* ($2n=24$) or species closely related to them. The genomes of the three species were designated as A_1 for *T. erecta*, B_1 for *T. tenuifolia*, and A_pB_p for *T. patula* by Towner (1961). The 24 chromosomes of *T. erecta* cv. Papaya Crush are mostly present in the form of 12 bivalents at meiosis. Univalents and quadrivalent were also observed in some cells (Chen and Lin 1981). The 48 chromosomes of *T. patula* are most frequently present as 24 bivalents. Multivalents were seen in 50.84% of the pollen mother cells (PMC) examined (Chen and Lin 1981). Analysis of chromosome pairing in the hybrids between *T. patula* and *T. erecta* could help explain the origin of *T. patula*. Bolz (1961), Jalil, Khoshoo and Pal (1974) reported that the 36 chromosomes of the hybrids between *T. patula* and *T. erecta* were mostly present as 12 bivalents and 12 univalents at meiosis. Trivalents were infrequently noted (Eresen 1966, Jalil *et al.* 1974). No other types of chromosome pairing in the hybrids have been reported. In this paper, we report 15 types of chromosome pairing in the hybrids in addition to 12 II+12 I. Pollen viability, seed fertility and meiotic irregularities observed in the hybrids are also reported. The origin of *T. patula* is considered.

Material and methods

The seeds of the three cultivars of interspecific hybrids between *T. patula* and *T. erecta* used in this study were obtained from W. Atlee Burpee Co. of Warminster, PA, USA. They were Gold Nugget, Orange Nugget, and Red and Gold Hybrid. PMC were used to study the chromosome pairing and the methods of preparing slides of PMC and measuring pollen viability have been described (Chen and Lin 1981).

Roman numerals are used in this report to represent valency, for example, I represents univalent, II represents bivalent, and so on. Standard errors were calculated for mean values.

Results

A wide variation of meiotic pairing occurred in the hybrids with various combinations of univalents (Figs. 1-3), bivalents (Figs. 1-3), trivalents (Fig. 2), and

Figs. 1-6. Meiosis in interspecific hybrids of *T. patula* and *T. erecta*. $\times 1280$. 1, 12 II+12 I at MI in Red and Gold Hybrid. 2, 9 II+12 I+2 III (arrows) in Red and Gold Hybrid. 3, 10 II+12 I+1 IV (arrow) in Red and Gold Hybrid. 4, a telophase II cell with many laggards in Orange Nugget. 5, a univalent lying away from the equatorial plane at MI in Red and Gold Hybrid. 6, three groups of chromosomes at TII in Orange Nugget.

quadrivalent (Fig. 3) (Table 1). Sixteen types of chromosome pairing were observed in 72 metaphase I (MI) cells and their frequencies are in Table 1. The 12 II+12 I association (Fig. 1) was the most frequent (30.56%, Table 1). An analysis of chromosome pairing revealed the presence of 6–14 univalents (mean 10.97 ± 0.20), 8–15 bivalents (mean 11.22 ± 0.15), 0–3 trivalents (mean 0.75 ± 0.10) and 0–1 quadrivalent (mean 0.08 ± 0.00) per cell (Table 2). A maximum of 15 bivalents occurred in a cell (Tables 1 and 2). About a half of the PMC (56.95%) analyzed con-

Table 1. Sixteen configurations of chromosome pairing at MI and their frequencies in interspecific hybrids of *Tagetes patula* and *T. erecta*

Chromosome pairing	Number of cells			Total	Frequency (%)
	Red and Gold Hybrid	Gold Nuggest	Orange Nuggest		
12 II+12 I	18	2	2	22	30.56
11 II+11 I+1 III	7	5	0	12	16.67
10 II+10 I+2 III	2	4	1	7	9.72
10 II+13 I+1 III	5	0	1	6	8.33
12 II+ 9 I+1 III	5	0	0	5	6.94
13 II+10 I	3	0	1	4	5.56
10 II+12 I+1 IV	2	0	1	3	4.17
11 II+ 8 I+2 III	0	1	1	2	2.78
11 II+14 I	2	0	0	2	2.78
9 II+ 9 I+3 III	1	1	0	2	2.78
14 II+ 8 I	1	1	0	2	2.78
9 II+12 I+2 III	1	0	0	1	1.39
15 II+ 6 I	1	0	0	1	1.39
10 II+ 6 I+2 III+1 IV	0	0	1	1	1.39
8 II+10 I+2 III+1 IV	0	0	1	1	1.39
8 II+13 I+1 III+1 IV	0	0	1	1	1.39
Total				72	

Table 2. Chromosome pairing in 72 MI cells of interspecific hybrids ($2n=36$) of *T. patula* and *T. erecta*

	Chromosome pairing (per cell)				% PMC having	
	I	II	III	IV	multivalents	univalents
Range (no.)	6–14	8–15	0–3	0–1	56.95	100.00
Mean no.	10.97	11.22	0.75	0.08		
Standard error	± 0.20	± 0.15	± 0.10	± 0.00		
*	30.84	62.34	6.25	0.92		

* Percentage of the total number of chromosomes associated in the given configuration per cell. e.g. for III, 0.75×3 chromosomes/36 chromosomes = 6.25%.

tained at least one multivalent (Tables 1 and 2). The trivalent appeared as a chain-of-3 (Fig. 2), a ring-and-rod, or a Y. Configurations of quadrivalent included chain-of-4 (Fig. 3) and ring-and-chain. No multivalents involving more than four chromosomes were seen. In Figs. 2 and 3 are cells with 9 II+12 I+2 III and 10 II+12 I+1 IV, respectively.

Lagging chromosomes occurred very frequently (Fig. 4, Table 3). The amount of cells with laggards among the three cultivars varied from 82.14 to 90.00% at MI and MII. The mean value was 83.33% at MI, 85.57% at MII and 84.28% at meiosis (Table 3). Among those cells with laggards, 35.43% had at least four laggards (Table 3). Very rarely, a cell may have a very large number of laggards (Fig. 4). Occasionally, chromosomes were off the equatorial plane (Fig. 5). Multiple aggregation of chromosomes (Fig. 6) and bridges were occasionally observed. The mean pollen viability of the three cultivars was 13.60%. The fertility of the seeds from Red and Gold Hybrid was 15.00%, but that of both Gold Nugget and Orange Nugget was 0% (Table 4).

Table 3. Meiotic irregularities in interspecific hybrids of *Tagetes patula* and *T. erecta*

Cultivar	Lagging						Bridging
	Meiosis I	Meiosis II	Meiosis I and Meiosis II	Total cells scored	No. of laggards	% cells with 4 or more laggards	Number of Bridges
Red and Gold Hybrid	15/17*	27/30	42/47	47	1-6		1
%	88.24	90.00	89.36			20.00	
Gold Nugget	49/59	42/50	91/109	109	1-9		1-4
%	83.05	84.00	83.49			34.80	
Orange Nugget	46/56	14/17	60/73	73	1-9		1-2
%	82.14	82.35	82.19			51.50	
Mean	83.33	85.57	84.28			35.43	

* Number of cells with laggards/total cells examined.

Table 4. Pollen viability and seed fertility in interspecific hybrids of *Tagetes patula* and *T. erecta*

Cultivar	Pollen* viability (%)	No. of viable pollen per 100	Seed fertility (%)
Red and Gold hybrid	14.73	7-23	15.00
Gold Nugget	12.46	7-20	0.00
Orange Nugget	13.62	4-23	0.00

* Mean value of three cultivates: 13.60%.

Discussion

If *T. patula* ($A_pA_pB_pB_p$) has two genomes contributed from *T. erecta* (A_1A_1) and the other two from a species remote to *T. erecta*, it can be assumed that in the hybrids of *T. patula* and *T. erecta*, chromosomes derived from *T. erecta* (A_1) would preferentially pair with one set of chromosomes from *T. patula* (A_p) giving 12 bivalents, and that the remaining chromosomes from *T. patula* (B_p) would be unpaired giving 12 univalents. Analysis of meiotic pairing in the hybrids showed that only about 30 percent of PMC exhibited the *Drosera* scheme. All cells examined had at least 6 univalents. Trivalents and quadrivalent were present in 56.95% of the PMC (Table 2). Eresen (1966) and Jalil *et al.* (1974) also reported

the presence of multivalents in the triploid hybrids, but only trivalents were found. Because about 10 percent (Table 1) of the cells analyzed had more than 12 bivalents, intragenomal pairing or reciprocal translocation in the genome from the unknown parental species can be assumed to have occurred. Bolz (1961) also observed a maximum of 15 bivalents in the species hybrids. If *T. erecta* and *T. tenuifolia* are truly the parental species of *T. patula*, bivalents may be expected to be present in monoloids of *T. tenuifolia* as a result of intragenomal pairing or reciprocal translocation.

Colchicine induced hexaploid *T. erecta-patula* ($2n=72$, $A_1A_1A_pA_pB_pB_p$) were, in average, found to have 33.95 bivalents, 1.1 univalents, and only 0.75 quadrivalent per cell. No trivalents were observed (Jalil *et al.* 1974). Thus A genomes in *T. erecta* (A_1A_1) and *T. patula* (A_pA_p) do not appear to correspond exactly because many more multivalents would have been expected. While absence of prevalent multivalents in a presumably polyploid species may be a result of increase in diploidization during the course of evolution (Gilles and Randolph 1951), diploidization could not be the cause for the low frequency of multivalents in this recently induced hexaploid. A low number of multivalents was also observed in induced polyploid plants of other species such as *Impatiens balsamina* (Raghuvanshi and Singh 1979), *Zinnia elegans* (Gupta and Koak 1976), *Trigonella foenum-graecum* L. (Raghuvanshi and Singh 1977) and *Portulaca grandiflora* (Singh 1979). Genetic factors (Riley and Chapman 1958, Feldman 1966) and small chromosome size could lead to a reduction in the number of multivalents. Therefore the absence of prevalent multivalents in the hexaploid *T. erecta-patula* cannot be regarded as an evidence against the proposal that *T. patula* originated from *T. erecta*.

On the other hand, *T. jaliscensis* ($2n=24$) is thought to be an alternate source of A genomes in *T. patula* (Towner 1962). It would be desirable to have synthetic tetraploids from *T. jaliscensis* and *T. tenuifolia* as well as triploids between *T. jaliscensis* and *T. patula*, and its corresponding hexaploids available so that studies can be made in hope to elucidate the origin of *T. patula*.

While pollen viability of both *T. erecta* and *T. patula* were approximately 25% (Chen and Lin 1981), the hybrids had a lower viability, 13.60%. There were at least six univalents in each PMC averaging 10.97 ± 0.20 per cell in the hybrids. The frequent occurrence of laggards (present in 84.29% of the PMC) can be correlated with the frequent occurrence of univalents. Physiological incompatibility and high frequency of laggards may be considered as factors contributing to the lower viability in these interspecific hybrids. Jalil *et al.* (1974) also reported a very low pollen viability of the triploid hybrids (0–33%) and 0 percent of seed fertility.

The three cultivars of triploid hybrids showed similar pollen viability; but Red and Gold Hybrid exhibited 15% seed fertility, Gold Nugget and Orange Nugget exhibited 0 percent. The differences in seed fertility were likely a result of genotypic differences among these cultivars. Intervarietal differences have been demonstrated for seed setting in some species. In polyploid *Phlox drummondii*, seed setting was highly variable ranging from 5% (White variety) to 95% (Yellow variety) (Raghuvanshi and Pathak 1975).

Summary

Chromosome pairing was studied in pollen mother cells (PMC) of the interspecific hybrids ($2n=36$) in *Tagetes patula* ($2n=48$) and *T. erecta* ($2n=24$). Among the 16 configurations of chromosomal pairing observed, the configuration with 12 bivalents plus 12 univalents was most frequently seen (30.56% of the PMC). A maximum of 15 bivalents was observed in a cell indicating some degree of intra-genomal pairing in the hybrids. Trivalents and quadrivalent were present in 56.95% of the PMC, and 7.17% of the 36 chromosomes was involved in multivalent formation. In average, there were 10.97 ± 0.20 univalents, 11.22 ± 0.15 bivalents, 0.75 ± 0.10 trivalent and 0.08 ± 0.00 quadrivalent per cell. Laggards were very frequently seen (84.28% of the PMC). Pollen viability was 13.6% partly due to meiotic irregularities. Seed fertility was very poor and physiological incompatibility may be considered a factor for the low fertility of the interspecific hybrids. The origin of *T. patula* is discussed.

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