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Analysis of meiosis in triticale (X*Triticosecale* Wittmack) × rye (*Secale cereale* L.) F_1 hybrids at three ploidy levels

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Summary. Triticales (X*Triticosecale* Wittmack) at three ploidy levels (8x, 6x, 4x, x = 7) were crossed with diploid rye (*Secale cereale* L.) to produce a solitary hypopentaploid hybrid (2n = 32), and a number of tetraploid (2n = 4x = 28) and triploid (2n = 3x = 21) hybrids. The hybrids exhibited a morphology which was intermediate between the parents. The number of bivalents ranged from 1–7 (4.65 per cell) in hypopentaploid, from 2–12 (7.13 per cell) in tetraploid and from 4–9 (6.84 per cell) in triploid hybrids. In 4x and 3x hybrids, trivalents and quadrivalents were also observed at low frequencies (range 0–1; mean 0.01–0.03 per cell). Chiasmata frequency was highest in triploid hybrids (12.44 per cell), lowest in hypopentaploid (5.37 per cell) and intermediate in tetraploids (10.54 per cell). More than 7^{11} were found in 39.7% pollen mother cells (PMC's) in the 4x hybrids and in 5.0% PMCs in 3x hybrids. It is concluded that an increase in the relative proportion of wheat chromosomes in the hybrids had a slight suppression effect on homologous as well as homoeologous pairing of rye chromosomes. Contrary to this, the relative increase in rye complement promoted homoeologous pairing between wheat chromosomes. In triploid hybrids, the chiasmata frequency as well as the 'c' value were the highest, suggesting that in tetraploid hybrids rye chromosomes had a reduced pairing (low frequency of ring bivalents).

Key words: Triticale × rye – Pairing regulation – Intergeneric hybrids

Introduction

In recent years, intergeneric hybrids in the tribe Triticeae have been produced in large number for improvement of cereal crops including wheat, triticale, barley and rye (Sharma and Gill 1983; Fedak 1985 a, b; Gupta and Fedak 1985 a, 1986 a).

In these intergeneric hybrids, analyses of meiosis gave useful information about the effect of individual genomes on pairing in hybrids. For instance, rye (*Secale cereale*) and other *Secale* species have been found to vary in their influence on meiotic pairing in various combinations (Gupta and Fedak 1985 a, b, c). It has also been shown that the effect of the *Secale* genome in hybrids with *Triticum* differ from that in hybrids with *Hordeum* (Gupta and Fedak 1985 b). The effect was found to depend on the dose of the rye genome relative to that of *Triticum* or *Hordeum*. In wheat-rye combinations, it had earlier been shown that while reduction of the dosage of wheat genomes increases the probability of homoeologous meiotic pairing between rye and wheat chromosomes, an increase in the dosage of rye genome increases the level of homoeologous pairing between wheat chromosomes (Miller and Riley 1972). Hybrids of triticale at three ploidy levels (8x, 6x, 4x) with diploid rye (2n = 2x = 14) provide interesting material for studying the effect of relative doses of wheat and rye genomes on meiotic pairing in triticale-rye or wheat-rye hbyrids.

In this communication, we present and discuss the results of meiotic analyses in hybrids (5x, 4x, 3x) produced by crossing triticales (X*Triticosecale*) at three ploidy levels (8x, 6x, 4x) with diploid (2x) rye (*Secale cereale*).

Materials and methods

Octoploid, hexaploid and tetraploid triticales were each crossed with diploid rye. The parents used and cross combinations made are presented in Table 1. The material for 8x triticales was obtained from the University of California, U.S.A.; that of 6x triticale from CIMMYT, Mexico; that of 4x

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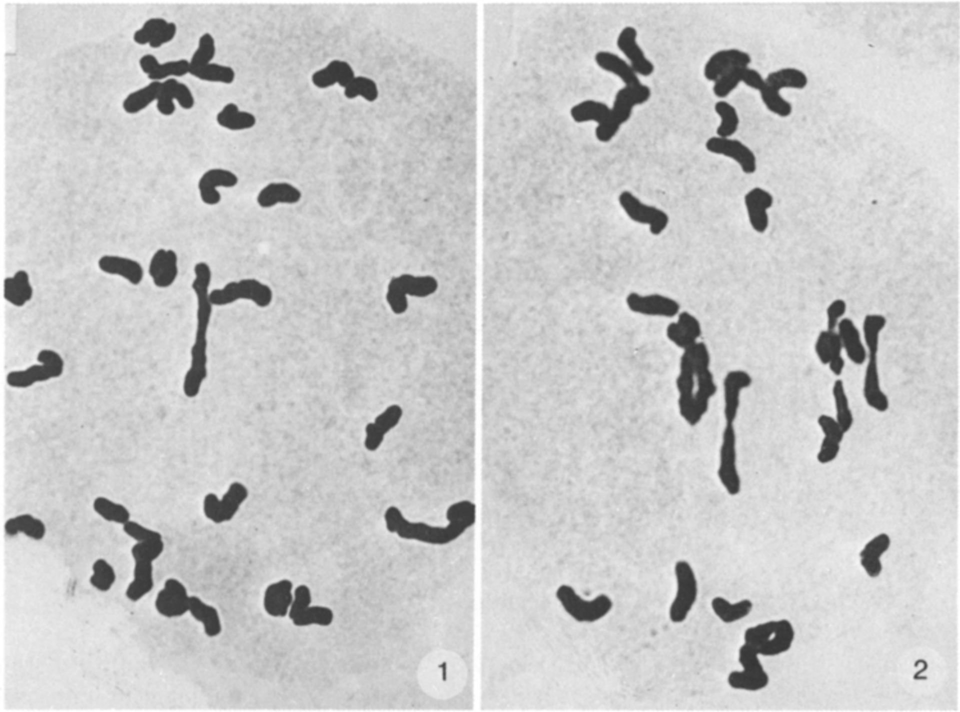


Figs. 1 and 2. Meiosis in a hypopentaploid (2n = 32) derived from a cross 8x triticale × 2x rye. **1** Metaphase I showing $30^{I} + 1^{II}$; **2** metaphase I showing $24^{I} + 4^{II}$ (3 rods + 1 ring)

Table 1. Cross combinations (triticale × rye) and parents used in the study (in every cross combination, the first parent is used as female)

Cross combination and parents	Chromosome no. in hybrid (2n)	Genomic constitution of hybrids
I. Combinations using 8x triticale (*AABBDDRR*)		*ABDRR*
1. 8x triticale (8A-94) × 2x rye (Assamese)	32	
II. Combinations using 6x triticale (*AABBRR*)		
2. 6x triticale (Armadillo) × 2x rye (Assamese)	28	*ABRR*
3. 6x triticale (6A 410) × 2x rye (Assamese)	28	*ABRR*
4. 6x triticale ('Beagle') × 2x rye (Assamese)	28	*ABRR*
5. 2x rye (Assamese) × 6x triticale (6A 410)	28	*ABRR*
III. Combinations using 4x triticale – $(A_xB_y)_2RR$[a]		
6. 4x triticale (80-03) × 2x rye (Assamese)	21	$(A_xB_y)RR$[a]

[a] $A_xB_y = 7$ chromosomes

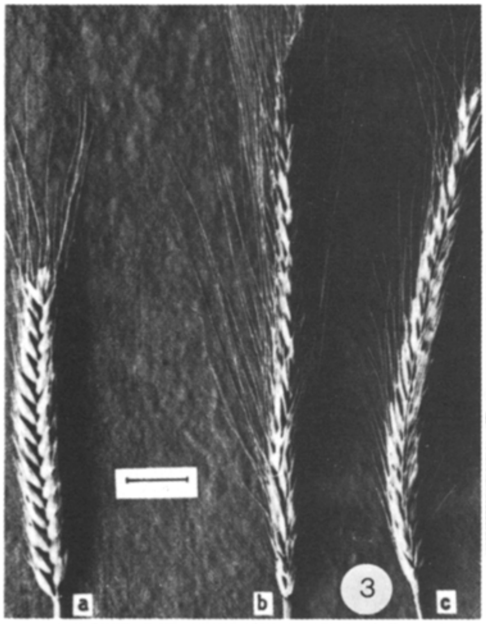
triticale from the University of Manitoba, Canada, and that of 2x rye from the National Bureau of Plant Genetic Resources (NBPGR), New Delhi. These were multiplied and maintained for several generations at Meerut University, Meerut, India. Crosses were made at Meerut University Experimental Farm and the F_1 hybrid plants were raised at the IARI Regional Station, Wellington (Tamil Nadu) in an off-season nursery.

The spikes of the F_1 plants were fixed in Carnoy's solution (6 : 3 : 1) for meiotic studies. A drop of $FeCl_3$ was added to the fixative as a mordant. The material was transferred to 70% ethanol after 24 h until squashes were made. Acetocarmine (2%) was used for staining. The relative affinities between genomes were calculated by using the mathematical procedures developed by Alonso and Kimber (1981) and Kimber and Alonso (1981).

Results

1 Meiotic analysis of hybrids

a) (8x triticale × 2x rye) hybrids (ABDRR). Only one F_1 plant survived in the cross between octoploid triticale



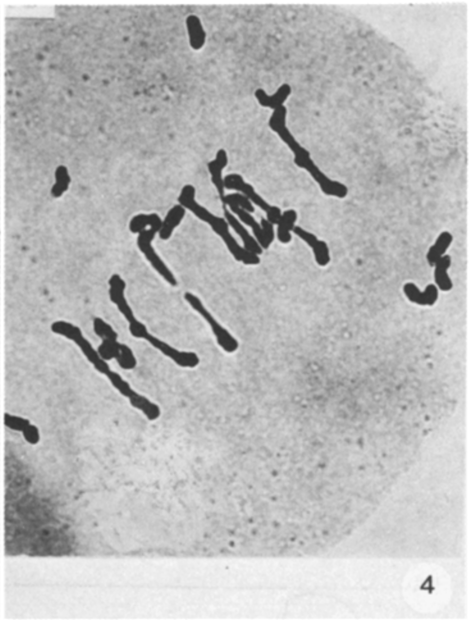


Fig. 3. Morphology of spikes of parent (**a** = triticale; **c** = rye) and the hybrid (**b**) of a cross 6x triticale × 2x rye

Figs. 4–6. Meiosis in hybrids ($2n = 4x = 28$) between 6x triticale and 2x rye. **4** Metaphase I showing $10^{I} + 9^{II}$ (7 rods + 2 rings); **5** metaphase I showing $14^{I} + 7^{II}$ (3 rods + 4 rings); Fig. **6** metaphase I showing $11^{I} + 7^{II}$ (2 rods + 5 rings) + 1^{III}

and rye. This exhibited a hypopentaploid chromosome number ($2n = 32$) instead of a euploid ($2n = 35$). At metaphase I, the number of bivalents ranged from 1 to 7 (Figs. 1 and 2; Table 2). Quadrivalents or heteromorphic bivalents were absent even though such abnormalities as anaphase bridges, misdivision of laggards and unequal distribution of chromosomes at anaphase I were noticed.

b) (6x triticale × 2x rye) hybrids (ABRR). The hybrids obtained from this cross had intermediate spike morphology with respect to the parents (Fig. 3). The data on chromosome associations and chiasmata frequencies are presented in Table 2. Most pollen mother cells (P.M.C.'s) exhibited 7 bivalents and 14 univalents as expected with an average bivalent frequency of 7.13 per P.M.C. In other cases, bivalents ranged from 2 to 12 (Figs. 4–6). Heteromorphic bivalents in a low frequency were also observed. Abnormalities such as formation of multivalents, bridges, unequal distribution of chromosomes, misdivision of laggards at anaphase I and formation of micronuclei were also noticed.

c) (4x triticale × 2x rye) hybrids (AxBy) RR. The hybrids obtained from this cross also had intermediate spike morphology with respect to their parents (Fig. 7). Most

Table 2. Chromosome associations and chiasmata frequencies in hypopentaploid (one plant), tetraploid (16 plants from four cross combinations) and triploid (one plant) hybrids of triticale and rye

Hybrid combination and genome constitution	No. of cells analyzed	II				III	IV	Chiasmata		% PMC's with	
		I	Ring	Rod	Total			Total	Per bivalent	7^{II}	more than 7^{II}
8x triticale × 2x rye (*ABDRR*; 2n = 32)	100	22.64 (18–28)[a]	0.44 (0–4)	4.21 (1–7)	4.65 (1–7)[a]	0.02 (0–1)	–	5.37 (1.20)[c]	1.15	8.0	0.00
6x triticale × 2x rye (*ABRR*; 2n = 28)	1,600	13.62 (6–22)	1.72 (0–6)	5.36 + 0.04[b] (0–12)	7.13 (2–12)	0.03 (0–1)	0.01 (0–1)	10.54 (2.17)[c]	1.48	25.50	39.70
4x triticale × 2x rye [(A_xB_y)*RR*; 2n = 21]	400	7.25 (3–11)	3.81 (1–7)	3.03 + 0.01[b] (0–5)	6.84 (4–9)	0.01 (0–1)	0.01 (0–1)	12.44 (1.85)[c]	1.82	74.50	5.00

[a] Values in parentheses are ranges

[b] Heteromorphic bivalents

[c] Interstitial chiasmata in parenthesis

Table 3. Relative genome affinity in triploid and tetraploid hybrids

Genome constitution of hybrid	Model giving best fit	C[a]	x[b]	Homoeologous chiasmata (%)	Ratio of homologous to homoeologous chiasmata
(A_xB_y)*RR* (2n = 21)	2:1	0.71	0.96	7.21	12.87:1
ABRR (2n = 28)	2:1:1	0.47	1.00	0.10	1024.00:1

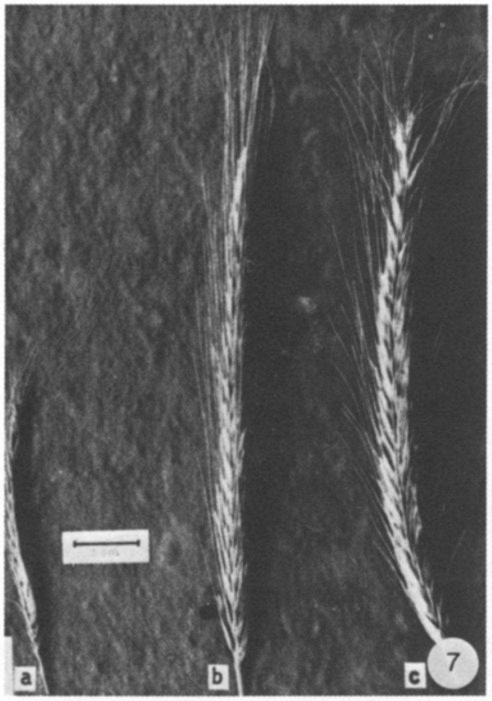
[a] Mean arm pairing frequency

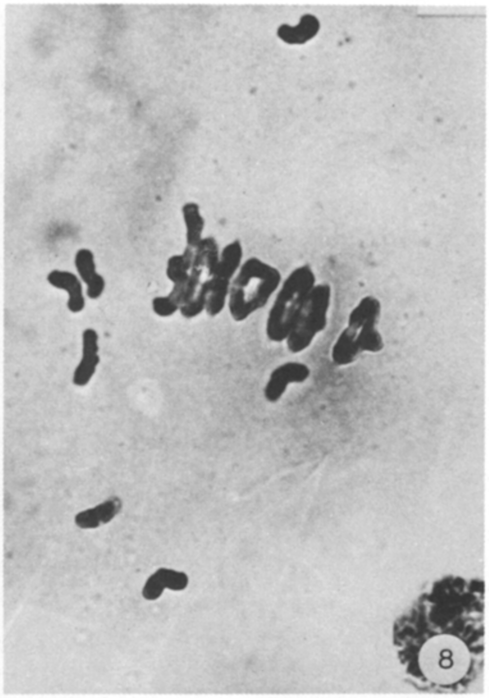
[b] Relative affinity of most closely related genomes

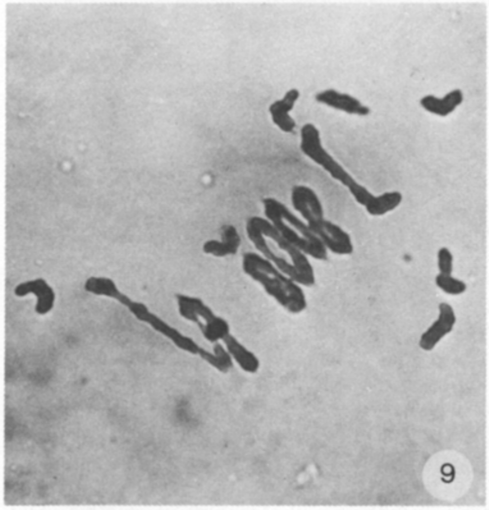
of the P.M.C.'s shared 7 bivalents and 7 univalents as expected (Fig. 8). The heteromorphic bivalents included only rod bivalents and were observed only in the cells with more than 7 bivalents. Quadrivalents were observed in a low frequency (Fig. 9). Separation of chromosomes at anaphase I was often regular (Fig. 10), although bridges and laggards were also observed in some cells.

Discussion

The hybrids produced and utilized during the present study, provide suitable material for a study of (a) the effect of the wheat genome(s) on pairing between rye homologues; (b) the effect of the rye genome on pairing between homoeologous chromosomes of wheat and (c) the effect of different combinations of wheat and rye genomes on pairing between homoeologous chromosomes of wheat and rye. In hybrids examined during the present study, three ploidy levels are represented and in each case seven pairs of rye chromosomes (six pairs when there is a substitution, e.g. *Armadillo*, Gustafson and Zillinsky 1973) are supposed to be present along with a variable number of non-homologous wheat chromosomes, and accompanied by the *Ph* locus to suppress homoeologous pairing. In such a situation, if rye chromosomes behave independently without being influenced by wheat chromosomes and if the wheat chromosomes do not pair either among themselves or with rye chromosomes, then one would expect seven bivalents (six in the case of *Armadillo*, due to substitution) in a large number of P.M.C.'s as in the normal diploid rye. A similar situation should be observed with respect to chiasma formation so that chiasma frequency in diploid rye and the above three hybrids should be the same, except when the triticale carried a substitution (e.g. *Armadillo*) and provided there are no interactions involved. Contrary to this oversimplified situation, we observed that the pairing behaviour and chiasmata frequencies varied in three cases and in one case it was much lower than what is known for diploid rye. For instance, in six inbreds of rye, the mean chiasmata frequency (averaged over at least six plants each) varied from 8.68 to 13.34 and in cultivar 'Petkuser Sommerroggen' (Ra 10), it was 13.69 (Lelley 1978). This sug-







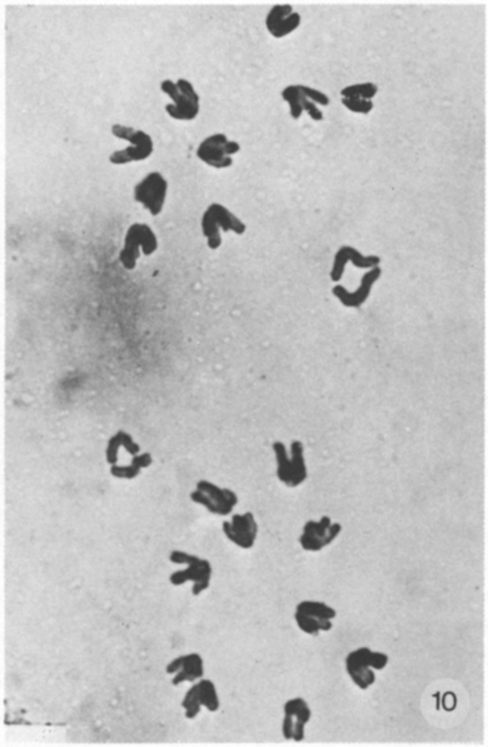


Fig. 7. Morphology of spikes of parents (**a** = triticale, **c** = rye) and the hybrid (**b**) of a cross, 4x triticale × 2x rye

Figs. 8–10. Meiosis in hybrids (2n = 3x = 21) between 4x triticale and 2x rye. **8** Metaphase I showing $7^{I}+7^{II}$ (1 rod + 6 rings); **9** metaphase I showing $7^{I}+5^{II}$ (1 rod + 4 rings) + 1^{IV}; **10** anaphase I showing a 10:9 distribution and two dividing univalents in a triploid hybrid (2n = 21) gests that the presence of wheat chromosomes influenced the pairing not only between rye chromosomes, but also between wheat and rye chromosomes.

The frequency of bivalents in tetraploids is higher than in triploids, although the chiasmata frequency is higher in triploids. Higher bivalent frequency in tetraploids may be attributed to pairing between chromosomes of two genomes of wheat and the pairing between wheat and rye chromosomes, as evident from the presence of multivalents and heteromorphic bivalents in triploid as well as tetraploid hybrids. The frequencies of mean ring bivalents, total bivalents and chiasmata were the lowest in the solitary hypopentaploid hybrid plant ($2n=32$).

The above results can be compared with earlier published results (Riley and Miller 1970; Miller and Riley 1972). Although the chiasmata frequencies suggest that an increase in the relative proportion of wheat chromosomes in hybrids had a slight suppression effect on homologous pairing of rye chromosomes, there is no definite evidence that a relative increase in rye chromosomes promoted homoeologous pairing. On the contrary, the number of cells with more than 7^{11} was higher in tetraploid hybrids than in triploid hybrids, suggesting that homoeologous pairing takes place mainly in tetraploid hybrids. It is interesting to note in a recent study that a different effect is observed when barley and rye genomes are brought together in hybrids (Gupta and Fedak 1985b). We also recognize that the two rye genomes in the hybrids belong to different cultivars, one of them from Assamese rye and the other from an unknown source present in triticale parent. This could also influence the chiasmata frequency, although this will be a common factor in all the three hybrids except that the three missing chromosomes in the pentaploid hybrid may or may not include one or more rye chromosomes. In such studies, it is difficult to distinguish the pairing configurations involving wheat and rye chromosomes from other possible configurations unless banding techniques or in situ hybridization techniques are utilized. There is, however, evidence available that homoeologous pairing between wheat and rye chromosomes is facilitated by a relatively higher proportion of rye chromosomes as evident from heteromorphic bivalents observed in tetraploid and triploid hybrids but not in the pentaploid hybrids. This may help transfer of rye segments to wheat through recombination.

The relative affinity tests demonstrated that the meiotic pairing follows 2 : 1 and 2 : 1 : 1 models in triploid and tetraploid hybrids, respectively. The relatively higher 'c' value in triploid hybrids would suggest reduced pairing of rye chromosomes in tetraploid hybrids and the difference in pairing can be attributed to the increased number of wheat chromosomes. On the other hand, in tetraploid hybrids, the bivalents formed due to homoeologous pairing may be more due to homoeologous chromosomes of A and B genomes which are supposed to be absent in triploid hybrids. The homoeologous pairing is higher in triploid hybrids as per mathematical analysis, where it takes place between wheat and rye chromosomes (Alonso and Kimber 1981). This confirms the recommendations that while introducing alien genetic variation, the number of wheat chromosomes should be kept minimum and rye chromosomes to the maximum (Miller and Riley 1972).

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References

Alonso LC, Kimber G (1981) The analysis of meiosis in hybrids. 2. Triploid hybrids. Can J Genet Cytol 23:221–234

Fedak G (1985 a) Alien species as sources of physiological traits for wheat improvement. Euphytica 34:673–680

Fedak G (1985 b) Wide crosses in *Hordeum*. In: Rasmusson D (ed) Barley monograph. Am Soc Agron, Monogr No 26:155–186

Gupta PK, Fedak G (1985a) Meiosis in seven intergeneric hybrids between *Hordeum* and *Secale*. Z Pflanzenzücht 95:262–273

Gupta PK, Fedak G (1985b) Genetic control of meiotic chromosome pairing in polyploids in the genus *Hordeum*. Can J Genet Cytol 27:515–530

Gupta PK, Fedak G (1985c) Variation in induction of homoeologous chromosome pairing in 6x *Aegilops crassa* by genomes of six different species of *Secale*. Can J Genet Cytol 27:531–537

Gupta PK, Fedak G (1986a) Wide crosses for improvement of wheats and barley. In: Proc 5th SABRAO Congr (in press)

Gupta PK, Fedak G (1986b) The inheritance of genetic variation in rye (*Secale cereale*) affecting homoeologous chromosome pairing in hybrids with bread wheat (*Triticum aestivum*). Can J Genet Cytol 28:844–851

Gustafson JP, Zillinsky FJ (1973) Identification of D-genome chromosomes from hexaploid wheat in a 42-chromosome triticale. In: Sears LMS, Sears ER (eds) Proc 4th Int Wheat Genet Symp. University of Missouri, Columbia MO, pp 225–232

Kimber G, Alonso LC (1981) The analysis of meiosis in hybrids. 3. Tetraploid hybrids. Can J Genet Cytol 23:235–254

Lelley T (1978) Genetic control of chiasma frequency and distribution in rye (*Secale cereale*). Can J Genet Cytol 20:471–474

Miller TE, Riley R (1972) Meiotic chromosome pairing in wheat-rye combinations. Genet Iber 24:241–250

Riley R, Miller TE (1970) Meiotic chromosome pairing in Triticale. Nature 227:82–83

Sharma HC, Gill BS (1983) Current status of wide hybridization in wheat. Euphytica 32:17–31