Theor. Appl. Genet. 64, 333–337 (1983)

Morphology and Chromosome Pairing of a Hybrid Between *Triticum durum* Desf. and *Haynaldia villosa* (L.) Schur.

A. Blanco, R. Simeone, O. A. Tanzarella
Institute of Plant Breeding, University of Bari (Italy)

B. Greco
Institute of Germplasm, C.N.R., Bari (Italy)

Summary. Intergeneric hybrids between *T. durum* Desf. ($2n=4x=28$, AABB) and *Haynaldia villosa* (L.) Schur. ($2n=2x=14$, VV) was obtained at a frequency of about 5.6% of pollinated florets. Phenotypically the F_1 plants resemble more the maternal parent than the *H. villosa* and are almost completely sterile. However, some seeds were obtained on selfed and backcrossed heads with the *durum* wheat parent. The hybrid had a somatic complement of $2n=3x=21$, ABV, with a mean chromosomal relationship of 13.62 univalents, 3.30 bivalents, and 0.26 trivalents. The high pairing was likely due to gene(s) of *H. villosa* interacting with the 5B homoeologous restricting system of wheat.

Key words: Intergeneric hybrids – *Triticum* – *Haynaldia* – Homoeologous pairing – Pairing promoters

---

Introduction

Interest in the interspecific hybridization in *Triticinae* has arisen from the need to exploit the variability of the related species for genetic improvement of *durum* and common wheat (McFadden and Sears 1947; Riley and Kimber 1966; Feldman 1979) and to investigate evolutionary relationships between species (Kihara 1954; Sears 1975; Alonso and Kimber 1981).

Haynaldia villosa (L.) Schur. is an allogamous annual plant native of the Mediterranean region; it is a diploid ($2n=14$) with a genome composition of VV. As a germplasm donor this species has been found to be interesting because of marked variability of some of its morphological characteristics, drought tolerance as well as resistance to disease such as powdery mildew, stem and leaf rusts (Hyde 1953; Pasquini et al. 1978; Scarascia et al. 1982).

Hybridization of *H. villosa* with diploid, tetraploid and hexaploid species of *Triticum* has been investigated (Strampelli cited in Raineri 1914; Sando 1935; Sears 1953; Kostoff and Arutinova 1937; McFadden and Sears 1947; Nakajima 1959; Halloran 1966; Gupta 1972; Meletti et al. 1977; Piralov 1981; Ibraginov 1979; Chen et al. 1982) and found to have different but usually low degrees of crossability (from 1.2 to 17.2% of pollinated florets). As in other instances of interspecific hybridization the crossability depends both on the specific genotypes involved and on the effects due to changes in seasonal conditions and crossing procedure. The *H. villosa* chromosomes have, none the less, been successfully added, collectively and individually, to the full chromosome complement of hexaploid wheat (Hyde 1953; Sears 1953).

This study was carried out to investigate the morphology, the level of chromosome pairing and the fertility of *T. durum* × *H. villosa* hybrids.

Materials and Methods

The *Triticum durum* Desf. ($2n=4x=28$, AABB) parent used in this study was the Italian semi-dwarf cultivar 'Creso'. The *H. villosa* (L.) Schur. ($2n=2x=14$, VV) strain was collected at the experimental farm of the Faculty of Agriculture, University of Bari (Italy), located 15 km from Bari.

The parental lines were grown in the field and during the spring of 1981; 250 *durum* wheat florets were emasculated and pollinated with *H. villosa.* Five embryos were excised from developing seeds 12–15 days after pollination and cultured on sterile B 5 medium (Gamborg et al. 1968). Of these, three embryos developed into plantlets on the medium and were transplanted into pots at the three leaf stage; two of them grew into vigorous F_1 hybrids. Ten other developing seeds were left to ripen on the plants and they yielded nine hybrid seeds. Although shrivelled, five of these germinated normally on moist filter paper in petri-dishes in the laboratory.

The root-tip of the F_1 hybrids were pre-treated for 4 h with 0.05% colchicine and then fixed in Carnoy's 3:1 fixative for mitotic studies. Individual anthers with PMC's at metaphase I were fixed in Carnoy's 6:3:1 fixative for meiotic studies. A standard Feulgen staining and squash procedure was used with both root-tips and PMC's to prepare material for cytological examination. Spike characteristics of the parental lines and F_1 hybrid were measured from 10 spikes.

Results and Discussions

The hybrid *T. durum* × *H. villosa* was produced with a relatively low frequency. In fact two seedlings were obtained from the five cultured embryos and five others were obtained from nine mature seeds. This represents a seed set of 5.6% and a hybrid seedling frequency of 2.8% of pollinated florets in the field conditions, since 250 pollinations were carried out. It is worth noting that the in vitro culture of the hybrid embryos excised from developing seeds 12–15 days after pollination is not needed to obtain hybrid plants, since most of the mature seeds had enough endosperm for a normal germination in petri-dishes.

All the plants analysed had the expected somatic chromosome number (2n = 21) and no chromosome instability was observed. Phenotypically the hybrid plants resemble the maternal parent more than the *H. villosa*. The hybrid spike characteristics are presented in Table 1, as are those for the *durum* wheat and *H. villosa* parents. The spikes of the hybrid (Fig. 1) were longer and with more spikelets than those of the *durum* wheat. Their glumes were provided with short awns, while the lemma awns were very long (about 10 cm). The rachis of the spike was as fragile and hairy as that of the *H. villosa*.

Table 1. Mean spike characteristics from 10 spikes of *T. durum* Desf. cv. "Creso", *T. durum* Desf. cv. "Creso" × *H. villosa* (L.) Schur. hybrid, and *H. villosa* (L.) Schur.

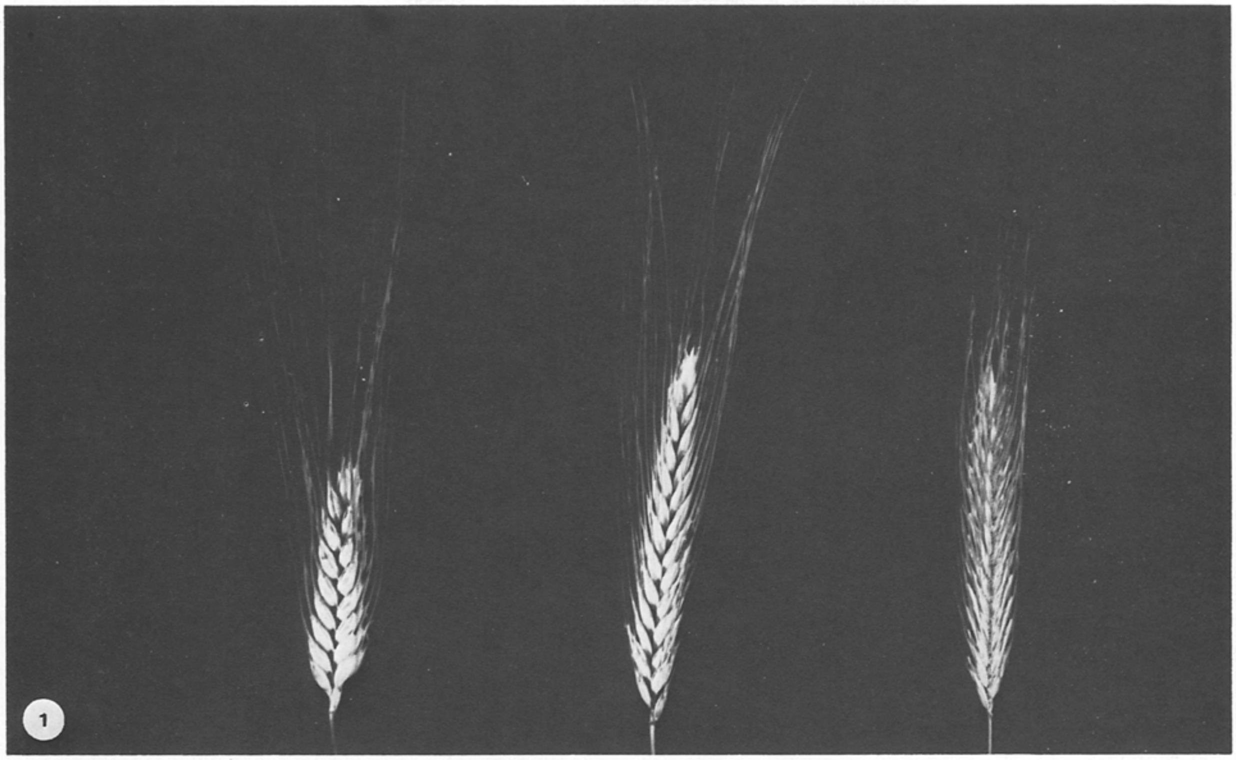
Character	*T. durum*	*T. durum* × *H. villosa*	*H. villosa*
Spike lenght, cm	6.1	9.2	8.4
Spike width, cm	1.6	1.4	1.5
Spikelet length, cm	1.7	1.9	2.0
Spikelet/spike, n.	16.6	23.4	25.1
Florets/spikelet, n.	4.8	5.0	4.2
Kernels/spikelet, n.	3.60	0.07	2.00
Pollen fertility, %	95.4	0.8	88.7
Glume body length, cm	1.1	1.0	0.7
Glume awn length, cm	0.1	1.4	3.7
Lemma body length, cm	1.1	1.3	1.3
Lemma awn length, cm	13.1	9.7	4.6
Anther length, mm	3.6	4.2	10.9
Glume hairiness	absent	absent	present
Rachis	tough	fragile	fragile

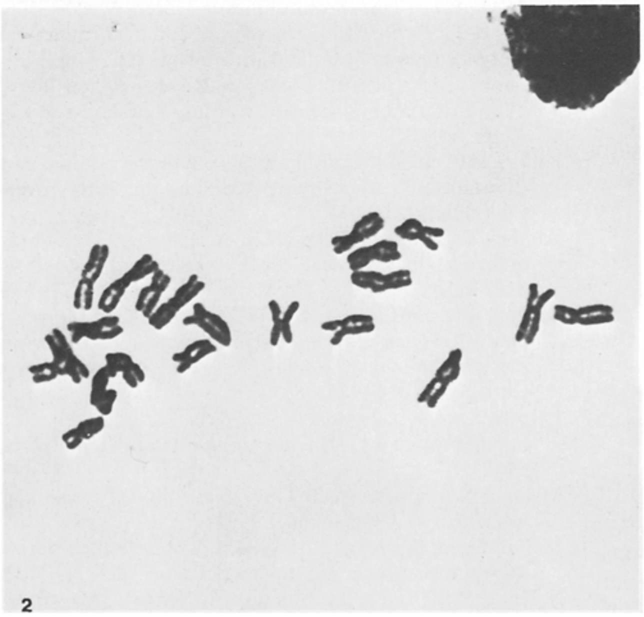
The pollen fertility, estimated as the percentage of pollen grains stainable with aceto-carmine, was less than 1% and the spikes were almost completely sterile; only 1 or 2 seeds per selfed and back-crossed heads with the *durum* wheat pollen were obtained. The chromosome number of the F_2 and BC_1 plants (2n = 42 and 2n = 35, respectively), along with the meiotic analysis of the PMC's of the F_1 plants (2n = 21), indicated that the fertility of the F_1's was very likely due to meiotic non reduction in a number of cells that resulted in male and female gametes with unreduced chromosome number. This process, that determines a certain fertility in interspecific and intergeneric hybrids expected to have little or no chromosome pairing, has been recently reviewed and demonstrated for *T. eldreichii* × *T. durum* F_1 hybrid by Mann and Sasakuma (1977).

Table 2 shows the results of the chromosome association at the first meiotic methaphase over 100 pollen mother cells of the F_1 hybrids *T. durum* × *H. villosa*, along with the chromosome pairing of the haploid of *T. durum* (Lacadena and Ramos 1968), and of the hybrids *T. dicoccoides* × *H. villosa* (McFadden and Sears 1947) and *T. aestivum* × *H. villosa* (euhaploid and nulli-5B haploid; Halloran 1966).

Table 2. Chromosome pairing at metaphase I in F_1 hybrids among *T. durum*, *T. dicoccoides*, *T. aestivum* and *H. villosa*, and in the haploid of *durum* wheat

Hybrid	Chromosome number 2n	No. of cells	Chromosome associations						References
			I	II			III	Other	
				Rod	Ring	Total			
T. durum × *H. villosa*	21	100	13.62 (8 – 19)	3.15 (1–6)	0.15 (0 – 2)	3.30 (1 – 6)	0.26 (0 – 2)	–	Authors
T. durum (euhaploid)	14	94	13.19	–	–	0.37	–	–	Lacadena and Ramos 1968
T. dicoccoides × *H. villosa*	21	100	19.35	–	–	0.79	–	0.07	McFadden and Sears 1947
T. aestivum × *H. villosa*	28	100	27.02	0.47	0.02	0.49	–	–	Halloran 1966
T. aestivum × *H. villosa*	27 (5B absent)	100	11.96	3.81	1.00	4.81	0.86	0.71	Halloran 1966





Figs. 1 – 3. 1 (left to right) *T. durum* cv "Creso"; *T. durum* × *H. villosa*; *H. villosa*. **2** Somatic metaphase of *T. durum* × *H. villosa* hybrid. **3** Meiotic metaphase in *T. durum* × *H. villosa* hybrid with $1^{III} + 6^{II} + 6^{I}$

The main chromosome pairing observed in the *durum* wheat × *Haynaldia* hybrid (3.15 rod and 0.15 ring bivalents, with a range from 1 to 6, and 0.26 trivalents, with a range from 0 to 2) was much higher than the haploid of *durum* wheat (Lacadena and Ramos 1968) which has an average of 0.37 bivalents per cell (with a range from 0 to 2 bivalents) due to autosyndesis within the wheat complement.

Previous studies on the association of *Haynaldia* and *Triticum* chromosomes at meiosis have indicated that the pairing is low and variable. McFadden and Sears (1947) observed 0.79 bivalents per cell in the

T. dicoccoides × *H. villosa* hybrid. In the crosses with *T. aestivum*, Halloran (1966) observed 0.49 bivalents, while Chen et al. (1982) detected 1.66 e 2.29 bivalents respectively in two hybrids having different wheat parents.

It is interesting to compare the meiotic behaviour of the *durum* × *H. villosa* hybrid with that of the *T. aestivum* × *H. villosa* nulli-5B haploid (Halloran 1966), considering that the latter has 7 chromosomes of the D genome. The pairing in the *durum* hybrid was only slightly lower than in that of the above cited hybrid which lacks the *Ph* gene (suppressor of homoeologous pairing) located on the long arm of the 5B chromosome (Sears and Okamoto 1958; Riley and Chapman 1958). The high pairing observed in the hybrid examined can be due to the presence of a gene (or genes) in the *H. villosa* genotype that interacts with the 5B homoeologous restricting system of wheat.

Genotypes that promote pairing or suppress the *Ph* effects have been found in *Ae. speltoides* (Riley et al. 1961), *Ae. mutica* (Riley and Law 1965) and *Ae. longissima* (Mello-Sampayo 1971). These promoters of pairing have no effect in the diploids themselves, where complete pairing of homologues is the rule and there are no homoeologues; however, in hybrids with wheat they increase the pairing potential of homoeologues. The level of pairing induced by the high-pairing *Ae. speltoides* and *Ae. mutica* strains is almost similar to the one taking place in the same hybrids in absence of chromosome 5B. Moreover, strains of these species were found which affected pairing less strongly or not at all (Dover and Riley 1972; Kimber and Athwal 1972; Dvorak 1972). Partial suppression of *Ph* evidently occurs in hybrids with tetraploid *Ae. caudata* (Riley 1968) and *Ae. umbellulata* (Riley et al. 1973). Mochizucki (1962), also, found that the addition of a particular pair of *Agropyron elongatum* chromosomes to *durum* wheat resulted in a substantial amount of homoeologous pairing.

If the *H. villosa* genotype suppresses the *Ph* effects, the pairing observed in the hybrid examined is presumed to be mainly between the A genome chromosomes with the corresponding homoeologues of genome B, such as in the nulli-5B haploid of common wheat (Riley and Law 1965) and in the hybrids of wheats lacking the *Ph* gene and rye (Dhaliwal et al. 1977; Giorgi and Cuozzo 1980). However, on the basis of trivalent frequency at meiosis in the *durum* wheat × *H. villosa* hybrid, it can be assumed that some chromosomes of *H. villosa* are pairing with those of *Triticum durum,* as already observed by Halloran (1966) in common wheat. The analysis of homoeologous pairing by Giemsa C-banding of the hybrid meiotic chromosomes can be establish the pairing degree between wheat and *Haynaldia* chromosomes. The fact that the *Haynaldia villosa* chromosomes can pair with those of wheat indicates that every useful character of *Haynaldia* can be transferred to the wheat complement by homoeologous recombination.

Literature

Alonso, L.C.; Kimber, G. (1981): The analysis of meiosis in hybrids. 2. Triploid hybrids. Can. J. Genet. Cytol. **23,** 271–283

Chen, S.Y.; Gao, L.Z.; Liu, M.H. (1982): A study of hybridization between *T. aestivum* and *Haynaldia villosa.* Plant Breed. Abstr. no. 4517

Dhaliwal, H.S.; Gill, B.S.; Waines, J. (1977): Analysis of induced homoeologous pairing in a *ph* mutant × rye hybrid. J. Hered. **68,** 206–209

Dover, G.; Riley, R. (1972): Variation at two loci affecting homoeologous meiotic chromosome pairing in *Triticum aestivum* × *Aegilops mutica* hybrids. Nature New Biol. **235,** 61–62

Dvorak, J. (1972): Genetic variability in *Aegilops speltoides* affecting homoeologous pairing in wheat. Can. J. Genet. Cytol. **14,** 371–380

Feldmann, M. (1979): Genetic resources of wild wheat and their use in breeding. In: Proc. Israeli-Italian Joint Meet. on Genet. Breed. Crop Plant, Rome

Gamborg, O.L.; Miller, R.A.; Ojima, K. (1968): Nutrient requirements of suspension cultures of soybean root cells. Exp. Cell Res. **50,** 151–158

Giorgi, B.; Cuozzo, L. (1980): Homoeologous pairing in a *ph* mutant of tetraploid wheat crossed with rye. Cereal Res. Commun. **3,** 485–490

Gupta, P.H. (1972): Cytogenetic evolution in the *Triticinae:* homoeologous relationships. Genetica **43,** 504–530

Halloran, G. (1966): Pairing between *Triticum aestivum* and *Haynaldia villosa* chromosomes. J. Hered. **57,** 233–325

Hyde, B.B. (1953): Addition of individual *Haynaldia villosa* chromosomes to hexaploid wheat. Am. J. Bot. **40,** 174–182

Ibraginov, M.K. (1979): Some results of work on the distant hybridization of *durum* wheat from Dagestan. Plant Breed. Abstr. no. 6069

Kihara, H. (1954): Consideration on the evolution and distribution of *Aegilops* species based on the analyser-method. Cytologia **19,** 336–357

Kimber, G.; Athwal, R.S. (1972): A reassessment of the course of evolution in wheat. Proc. Natl. Acad. Sci. USA **69,** 912–915

Kostoff, D.; Arutiunova, N. (1937): Studies on the polyploid plants *Triticum-Haynaldia* hybrids with special reference to the amphiploids *Triticum dicoccum* × *Haynaldia villosa.* Curr. Sci. **5,** 414–415

Lacadena, J.R.; Ramos, S. (1968): Meiotic behaviour in haploid plant of *Triticum durum* Desf. Genet. Iber. **20,** 55–71

Maan, S.S.; Sasakuma, T. (1977): Fertility of amphihaploids in *Triticinae.* J. Hered. **68,** 87–94

McFadden, E.S.; Sears, E.R. (1947): The genome approach in radical wheat breeding. J. Am. Soc. Agr. **39,** 1011–1026

Meletti, P.; Onnis, A. Stefani, A. (1977): *Haynaldoticum sardoum* Meletti et Onnis e la sua origine. G. Bot. Ital. **111,** 376

Mello-Sampayo, T. (1971): Promotion of homoeologous pairing in hybrids of *Triticum aestivum* × *Aegilops longissima.* Genet. Iber. **23,** 1–9

Mochizuki, A. (1962): *Agropyron* addition lines of *durum* wheat. Seiken Jiho **13,** 133–138

Nakajima, G. (1959): Cytogenetical studies of trigeneric hybrids. 2. Somatic chromosome numbers, external morphology and fertility of *Tper* HRF_1 and *Tdur* HRF_1. Jpn. J. Genet. **34,** 61–69

Pasquini, M.; De Los Angeles Gicas, M.; Vallega, J. (1978): *H. villosa* (L.) Schur. come fonte di resistenza alle ruggini e all'oidio, da incorporare nelle specie di frumento duro coltivate. Atti Giornate Fitopatologiche 349–353

Piralov, G.R. (1981): Two year's results from hybridizing wheat with *Haynaldia villosa* Schur. Plant Breed. Abstr. no. 10300

Raineri, L. (1914): La stazione di granicoltura di Rieti. Italia Agr. **51,** 6–12

Riley, R.; Chapman, V. (1958): Genetic control of the cytologically diploid behaviour of hexaploid wheat. Nature **182,** 713–715

Riley, R.; Kimber, G.; Chapman, V. (1961): Origin of genetic control of diploid-like behaviour of polyploid wheat. J. Hered. **52,** 22–25

Riley, R.; Law, C.N. (1965): Genetic variation in chromosome pairing. Adv. Genet. **13,** 57–114

Riley, R.; Kimber, G. (1966): The transfer of alien genetic variation to wheat. Ann. Rep. Plant Breed. Inst., Cambridge 1964–1965, 6–36

Riley, R. (1968): The basic and applied genetics of chromosome pairing. In: Proc. 3rd Int. Wheat Genet. Symp., pp. 185–195. Canberra: Aust. Acad. Sci.

Riley, R.; Chapman, V.; Miller, T.E. (1973): The determination of meiotic chromosome pairing. In: Proc. 4th Int. Wheat Genet. Symp. (eds. Sears E.R.; Sears, L.M.S.), pp. 731–738. Columbia, Missouri: Mo. Agr. Exp. Stn.

Sando, W.J. (1935): Intergeneric hybrids of *Triticum* and *Secale* with *Haynaldia villosa.* J. Agric. Res. **51,** 759–800

Scarascia Mugnozza, G.T.; De Pace, C.; Tanzarella, O.A. (1982): *Haynaldia villosa* (L.) Schur.: una specie di potenziale valore per il miglioramento genetico del frumento. I. Analisi di alcuni caratteri morfologici. Genet. Agrar. **36,** 76–77

Sears, E.R. (1953): Addition of the genome of *Haynaldia villosa* to *Triticum aestivum.* Am. J. Bot. **40,** 168–174

Sears, E.R.; Okamoto, M. (1958): Intergenomic chromosome relationships in hexaploid wheat. Proc. 10th Int. Congr. Genet. **2,** 258–259

Sears, E.R. (1975): The wheats and their relatives. In: Handbook of genetics (eds. King, R.C.), vol. 2, pp. 59–91. New York: Plenum Press

Received December 1, 1982
Communicated by R. Riley

Dr. A. Blanco
Dr. R. Simeone
Dr. O.A. Tanzarella
Institute of Plant Breeding
University of Bari
Via Amendola, 165/A
Bari (Italy)

Dr. B. Greco
Institute of Germplasm
Via Amendola, 165/A
Bari (Italy)