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## I. Research Notes

### Cytogenetics of branched spike in bread wheat

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Utilising chromosome 5B manipulations, JOSHI & SINGH (1979), have been able to transfer several desirable traits from rye into bread wheat. One single cross of hexaploid wheat variety PbC591 × ryes has generated enormous variability. A line from this material was segregating for normal and branched ear heads. It was observed that there were 5 plants which had branched ear-heads. The plants having branched spikes as well as the plants possessing normal spikes were picked up for further study.

All the spikes of these plants were selfed. Spikes were taken from branched as well as normal plants for meiotic studies. Observations at first meiotic metaphase revealed that all the plants producing branched ear-heads were trisomics ( $2n=43$ ) and normal looking plants were disomics ( $2n=42$ ). A regular trivalent was observed in a large number of cells of ramificated spikes while meiosis in disomics showed regular 21 bivalents.

Seeds obtained from branched and normal spikes, after selfing, were sown as spike progenies in the field to study the breeding behaviour of ramification. Normal spiked progenies produced only normal type of plants. Progenies of all the branched spikes segregated for branched type as well as normal type of spikes. A total of 253 plants were observed. Out of these, 230 were normal spiked while 23 were branched type. Meiotically all the branched plants were found to carry 43 chromosomes with a trivalent and normal plants with 42 chromosomes showed regular 21 bivalents.

Spikelet number in normal spikes ranged from 23 to 25 while in branched spikes the range observed was 75 to 87. Ramificated plants were 60–70 cms tall and normal plants were 90 to 100 cms tall.

Branching in the spikes of *Triticum* has been reported in derivatives of inter-varietal crosses by KORIC (1967, 1971, 1974, 1978), interspecific and intergeneric crosses by SHARMAN (1944),

TSITSIN (1965) and as induced mutations by SWAMINATHAN *et al.* (1966). Occurrence of ramificated spikes of *Triticum aestivum* Var. NP797 in an  $M_2$  population might have resulted from a deletion as suggested by SWAMINATHAN *et al.* (1966). KORIC (1978) proposed two genes, ramifera (*Rm*) and tetrastichon (*Ts*) for ramification. He suggested that ramification is the product of complementary gene action of *Rm* and *Ts* genes. On the basis of meiotic studies of ramificated spikes ( $2n=43$ ), which showed a trivalent in majority of the cells, it is suggested that ramification in *Triticum aestivum* in the present case may be as a result of additional gene dosage due to trisomic condition. Identification of the chromosomes involved in the trivalent is in progress.

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## Development of monosomic lines in durum wheat\*

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The cultivated species of wheat, *Triticum durum* ( $n=14:AABB$ ) and *T. aestivum* ( $n=21:AABBDD$ ) behave meiotically as true allopolyploids, with regular bivalent formation inspite of intergenomic pairing ability that exists between homeologous chromosomes. This meiotic behaviour which confers the tetraploid and hexaploid wheats a great cytological stability and high fertility is controlled by one or more genes which inhibit pairing of homeology chromosomes (OKAMOTO 1957; RILEY 1958; 1960; RILEY & CHAPMAN 1958).

Genetic analysis in polyploid wheats have been difficult due to duplication or triplication of genes and aneuploidy (SEARS 1954) has become one of the best tools in genetic and cytogenetic analysis of polyploid wheats. Attempts to obtain monosomics in tetraploid wheats have not been successful since such individuals are very weak and sterile having a low transmission ability of the monosomic condition (MOCHIZUKI 1968 a, b). The present investigation deals with the possibilities of isolating monosomic lines in *durum* wheat cv Bijaga Yellow by chromosome conversent method.

### Material and Methods

To obtain monosomic lines in *T. durum* cv 'Bijaga Yellow', the first fourteen monosomic lines of Pb C 591 (Monosomic for A- or B-genomes) were crossed to Bijaga Yellow. In  $F_1$  generation, the majority of the populations were of two types i.e. plants with 34 chromosomes ( $13''+8'$ ) which are monopentaploids and the second type being the plants with 35 chromosomes ( $14''+7'$ ) which are eupentaploids. In  $F_1$  generation the plants with 34 chromosomes were selected and back crossed to Bijaga Yellow. In first back cross generation, plants with  $13''+2'$  were selected and were again back crossed to Bijaga Yellow to get monosomic plants of the donor parent. In second back cross generation the plants with only  $13''+1'$ , were selected and back crossed to Bijaga Yellow to raise  $BC_3$  generation. During the course of back crossing the chromosomes of the D-genome were eliminated.

### Results and Discussion

The utility of monosomics and nullisomics in cytogenetic studies depends upon their breeding behaviour. Theoretically, one expects an individual with  $2n-1$  chromosomes to produce 'n' and 'n-1' gametes in equal frequency. Like wise one should be able to

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\* Part of the Ph. D. thesis submitted by the senior author to the University of Agricultural Sciences, Bangalore, India.

obtain '2n' and '2n-1' individuals in equal frequency from  $(2n-1) \times 2n$  crosses. Further more,  $2n-1$  individuals when selfed should yield  $2n$ ,  $2n-1$  and  $2n-2$  progeny in the ratio of 1:2:1. However, this is never realized in many cases and the departure from this expected ratio is indeed varied. The causes for this departure from expectation are: 1) production of 'n' and 'n-1' spores in unequal frequency; 2) reduced viability of 'n-1' spores; 3) competition between 'n' and 'n-1' microspores; 4) reduced viability of ' $2n-1$ ' zygotes; and 5) reduced viability of nullisomic zygotes. These factors mainly determine the breeding behaviour of the monosomics.

Table 1. Frequency of plants with different chromosome configurations at metaphase-I of meiosis in monopentaploid hybrids of crosses between first fourteen monosomic lines of Pb C 591 (monosomic for A- and B-genomes) and Bijaga Yellow

Populations	Total No. of plants analysed	Number of plants with 34 chromosomes	Number of plants with 35 chromosomes	Others
1A	20	11 (55.00)*	9 (45.00)	—
2A	20	8 (40.00)	10 (50.00)	2 (10.00)
3A	19	9 (47.37)	8 (42.11)	2 (10.53)
4A	16	10 (62.50)	6 (37.50)	—
5A	19	14 (73.68)	5 (26.32)	—
6A	20	12 (60.00)	7 (35.00)	1 (5.00)
7A	20	13 (65.00)	6 (30.00)	1 (5.00)
1B	15	10 (66.60)	5 (33.33)	—
2B	22	15 (68.18)	7 (31.81)	—
3B	20	11 (55.00)	8 (40.00)	1 (5.00)
4B	20	13 (65.00)	7 (35.00)	—
5B	15	9 (60.00)	6 (40.00)	—
6B	22	12 (54.55)	8 (36.36)	2 (9.09)
7B	16	10 (62.50)	5 (31.25)	1 (16.25)
Total:	249	148 (59.44)	91 (36.54)	10 (4.02)

\* Values in the parenthesis indicate the percentages.

The female parent, i.e. monosomic line Pb C 591 may produce either 'n' or 'n-1' gametes in different proportions. It is known that the transmission of monosomic condition from female side is 75 per cent. Thus when monosomic lines of Pb C 591 were used as female parent and the tetraploid wheat variety Bijaga Yellow as male parent 75 percent of monopentaploid plants were expected in F<sub>1</sub> generation. However, on an average 59.44 percent of the plants were monopentaploids (13''+8'), 36.54 percent eupentaploids and in the remaining 4.02 percent plants, the chromosome number was 2n=33 (Table 1). The occurrence of monopentaploids was maximum in line 5A, and in line 2A it was minimum.

Table 2. Frequency of plants with various chromosome configurations at metaphase-I of meiosis of monopentaploid (13''+8') lines (monosomic for A or B genome) × Bijaga Yellow in first back cross generation

Line (1)	Plants with different chromosome numbers								Total No. of plants analysed (10)
	28 chromosomes		29	30	31	32	33	34	
	13''+2' (2)	14'' (3)	13''+3' (4)	15''+4' (5)	13''+5' (6)	13''+6' (7)	13''+7' (8)	13''+8' (9)	
1A	2 (10.53)*	2 (10.53)	3 (15.79)	4 (21.04)	3 (15.79)	2 (10.53)	—	3 (15.79)	19
2A	1 (6.67)	3 (20.00)	1 (6.67)	3 (20.00)	1 (6.67)	4 (26.67)	—	2 (13.32)	15
3A	2 (12.50)	3 (18.75)	2 (12.50)	1 (6.25)	3 (18.75)	1 (6.25)	—	2 (12.50)	16
4A	2 (4.35)	2 (8.70)	3 (13.04)	5 (21.74)	3 (13.04)	3 (13.04)	2 (12.50)	4 (17.39)	23
5A	3 (15.00)	5 (25.00)	2 (10.00)	4 (20.00)	—	4 (20.00)	2 (8.70)	2 (10.00)	20
6A	1 (11.11)	3 (33.34)	1 (11.11)	2 (22.22)	—	—	—	2 (22.12)	9
7A	1 (7.14)	2 (14.29)	2 (14.29)	1 (7.14)	4 (28.56)	2 (14.29)	—	2 (14.29)	14
1B	1 (7.69)	3 (23.08)	3 (23.08)	4 (30.77)	—	—	—	2 (15.38)	13
2B	2 (13.33)	2 (13.33)	2 (13.33)	3 (20.00)	1 (6.68)	2 (13.32)	—	3 (20.00)	15
3B	2 (10.00)	4 (20.00)	4 (20.00)	6 (30.00)	—	—	—	4 (20.00)	20
4B	1 (7.14)	3 (21.42)	1 (7.14)	2 (14.29)	2 (14.29)	2 (14.29)	1 (7.14)	2 (14.29)	14
5B	—	2 (50.00)	1 (25.00)	1 (25.00)	—	—	—	—	4
6B	2 (16.67)	3 (29.00)	2 (16.67)	2 (16.67)	1 (8.33)	1 (8.33)	—	1 (8.33)	12
7B	1 (12.50)	1 (12.50)	2 (25.00)	1 (12.50)	1 (12.25)	—	—	2 (25.00)	8
Total:	20 (9.90)	18 (18.80)	29 (14.36)	39 (19.30)	19 (9.41)	21 (10.40)	5 (2.48)	13 (15.35)	202

\* Values in the parenthesis indicate the percentage.

Table 3. Frequency of plants with various chromosome configurations at metaphase-I of meiosis in crosses between plants with 13''+2' and Bijaga Yellow in second back cross generation

Line	Plants with different chromosome numbers					Total No of plants analysed
	27	28	29	30	31	
1A	1 (1.69)*	36 (61.03)	18 (30.51)	3 (5.08)	1 (1.69)	59
2A	—	16 (88.88)	1 (5.56)	1 (5.56)	—	18
3A	1 (2.63)	29 (76.32)	7 (18.42)	1 (2.63)	—	38
4A	1 (2.78)	28 (77.78)	6 (16.66)	1 (2.78)	—	36
5A	—	61 (83.56)	1 (15.07)	1 (1.37)	—	73
6A	2 (4.08)	33 (67.35)	12 (24.49)	2 (4.08)	—	49
7A	1 (2.32)	29 (87.44)	10 (23.26)	2 (4.66)	1 (2.32)	43
1B	—	31 (72.10)	10 (23.26)	1 (2.33)	1 (2.33)	43
2B	1 (2.00)	41 (82.00)	8 (16.00)	—	—	50
3B	1 (2.75)	36 (63.17)	18 (31.58)	1 (1.75)	1 (1.75)	57
4B	1 (2.38)	31 (73.81)	10 (23.81)	—	—	42
5B	2 (4.55)	32 (72.73)	9 (20.45)	1 (2.27)	—	44
7B	—	28 (70.00)	10 (25.00)	1 (2.50)	1 (2.50)	40
Total	11 (1.86)	431 (72.81)	130 (21.96)	15 (2.53)	5 (0.84)	592

\* Values in the parenthesis indicate the percentages.

In first back cross generation (BC<sub>1</sub>) plants with chromosome configurations from 13''+2' to 13''+8' occurred (Table 2). But plants with 13''+1, were observed, which agrees with, MOCHIZUKI (1968b). In the BC<sub>1</sub> generation, about 9.90 percent of the populations had 13''+2', 18.80 percent had 14'', 14.36 percent had 13''+5', 10.40 percent had 13''+6', 2.48 percent had 13''+7' and 15.25 percent had 13''+8'. The frequency of plants with 14 bivalents in the BC<sub>1</sub> generation, suggested that increasing homozygosity of the tetraploid complement in monopentaploid wheat hybrid reduced the frequency of univalent chromosome transmission. The low frequency of 13''+2' differed from MOCHIZUKI (1968b) who obtained about 30 to 50 percent plants with 13''+2'.

In the second back cross generation (BC<sub>2</sub>), the chromosome number varied from 27 to 31 (Table 3). Plants with 27 chromosomes occurred with least frequency (1.86 percent),



whereas plants with 14 bivalents occurred in highest frequency (72.81 percent). Frequency of monosomic plants in the second back cross progenies varied from zero to 4.55 percent. MOCHIZUKI (1968b) however observed only 0.33 to 7.40 percent monosomic plants in second back cross generation. The results of the present study indicate that the occurrence of monosomic plants in tetraploid wheat is quite low in contrast to that obtained in hexaploids. Therefore, the maintenance of the monosomic lines of tetraploid wheat requires laborious work. Moreover, the seeds obtained were also shrivelled, indicating poor endosperm development occurred in plants which are deficient for a single chromosome in tetraploid wheat Bijaga Yellow.

Table 4. Frequency of plants with monosomic ( $13''+1'$ ) and disomic ( $14''$ ) chromosome configurations lines ( $13''+1'$ )  $\times$  Bijaga Yellow in third backcross generation

Line	Frequencies of plants with		Total number of plants analysed
	$13''+1'$	$14''$	
1A	1 (2.17)*	45 (97.83)	46
3A	1 (1.89)	53 (98.11)	54
4A	—	39 (100.00)	39
6A	1 (1.89)	52 (98.11)	53
7A	—	36 (100.00)	36
2B	1 (2.33)	34 (97.67)	44
4B	—	43 (100.00)	43
6B	2 (4.55)	42 (95.45)	44
Total	6 (1.68)	352 (98.32)	358

\* Values in the parenthesis indicate the percentage.

When monosomic plants were back crossed to Bijaga Yellow, in the  $BC_3$  generation, the frequency of transmission of monosomic condition varied from zero to 4.55 percent (Table 4). On an average, the transmission frequency of monosomic condition was 1.68 per cent. About 98.32 percent plants obtained in the  $BC_3$  generation had 14 bivalents. KIHARA & TSUNEWAKI (1962), TSUNEWAKI (1964a, b) and LACADENA (1973) observed zero percent transmission frequency of mutagenic induced monosomic lines in *durum* wheat.

The low transmission rate of monosomic condition in tetraploid wheats may be due to selection against gametes or zygotes which are deficient in chromosomes.

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## Hybrid seed set in wheat × *Aegilops* crosses

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The wild relatives of wheat represent a valuable source of genetic variability for wheat breeding and *Aegilops* species are the richest source of resistance among wheat relatives. Several attempts have been made to transfer desirable chromosomal elements from *Aegilops* species to wheat. However difficulties in obtaining F<sub>1</sub> hybrids, low germination in hybrid seeds and sterility of F<sub>1</sub> hybrids are main barriers in wheat × *Aegilops* breeding programmes.

In this study four commercially grown common wheat cultivars (Yayla 305, 4/11, Köse 220/39, Sürak 1593/51) and four durum wheat cultivars (Beyaziye, Çakmak 79, L-51, S. Bursa 7113) were crossed as female parents with seven different *Aegilops* species as shown in Table 1, 2. All *Aegilops* species were collected from native rangelands in different regions of Turkey, except *Ae. squarrosa* which was introduced. The parents were seeded on experimental plots at three different dates in fall 1979 in order to synchronize the flowering periods.

Emasculation and pollination studies were made in the early morning between 7.00 to 10.00 A.M. emerging spikes of wheat were selected for emasculation. The uppermost and lowermost two spikelets were removed. Approximately one-third part of the glumes in spikelets left on the spikes were cut. The primary florets in each spikelet were emasculated and secondary floret in the middle was removed. Emasculated spikes were bagged with parchment bags and checked in the next morning. When the stigmas were fully receptive, mature anthers were taken from *Aegilops* just before the bursting of the pollen sacs, and at least one anther was carefully placed on each stigma in the wheat spikes. Pollinated spikes were bagged again. The spikes were harvested at maturity. Hybrid seed set was expressed as the percentage of hybrid seed to number of the florets emasculated, in each combination.

Two months later all hybrid seeds were placed between the layers of blotter papers for a germination test under laboratory conditions. Germination percentages of hybrid seeds were recorded and root tips were excised. Chromosome counts, using root tips, were made for all seeds.

Low percentages of hybrid seed set have frequently been recorded in wheat × *Aegilops* crossing studies (DOSBA & CAUDERON 1972; GOTSOV & PANAYOTOV 1972; VARDI 1973; KASCHIRI 1974; MAMMADOV 1976). In our studies, however, clearly higher seed set was obtained (Table 1, 2) and chromosome countings also verified that all seeds were hybrid. The mean seed sets were 18.4% and 23.9% respectively in common wheat × *Aegilops* and durum wheat × *Aegilops* crosses. The highest seed set values were obtained in common

Table 1. Seed set and germinability in *T. aestivum* × *Aegilops* crosses

Crosses*	No. of floret pollinated	Seed set		Germinability
		Number	%	
<i>T. aest.</i> × <i>Ae. triuncialis</i>	713	231	32.4	26.8
× <i>Ae. triaristata</i>	712	114	16.0	75.4
× <i>Ae. biuncialis</i>	457	114	24.9	100.0
× <i>Ae. umbellulata</i>	733	59	8.0	79.7
× <i>Ae. speltoides</i>	236	26	11.0	92.3
× <i>Ae. squarrosa</i>	150	9	6.0	100.0
Total	3001	553	18.4	61.8

\* *T. aestivum* × *Ae. ligustica* crosses were not made since flowering time of the parents could not be synchronized.

Table 2. Seed set and germinability in *T. durum* × *Aegilops* crosses

Crosses	No. of floret pollinated	Seed set		Germinability
		Number	%	
<i>T. durum</i> × <i>Ae. triuncialis</i>	1402	493	35.2	2.0
× <i>Ae. triaristata</i>	916	233	25.4	7.3
× <i>Ae. biuncialis</i>	714	156	21.8	39.7
× <i>Ae. umbellulata</i>	886	99	11.2	85.9
× <i>Ae. speltoides</i>	404	62	15.3	33.9
× <i>Ae. ligustica</i>	98	15	15.3	86.7
× <i>Ae. squarrosa</i>	90	1	1.1	—*
Total	4510	1059	23.5	19.6

\* Because of limited number of hybrid seed germinability value could not be calculated.

and durum wheat × *Ae. triuncialis* combinations.

In former studies on wheat × *Aegilops* hybrids low, sometimes no germination was found (KASCHIRI 1974; KOZHAKMETOV & ERLEPESOV 1974). The germination percentages of seeds varied greatly among the combinations; e.g very high germination, up to 100% was obtained in some crosses, while some crosses exhibited very low percentages (Table 1, 2).

Probably the main reasons for high seed set in wheat × *Aegilops*, were the crossing technique used and particularly, the climatic conditions prevailing during the crossing studies. The mean relative humidity and temperature in this period were 67% and 15°C, respectively. It was concluded that ample seed set in wheat × *Aegilops* crosses can be expected using a proper crossing technique under convenient humidity and temperature conditions.

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## Telocentric mapping of the squarehead (*vulgare*) gene *Q* on chromosome 5A of hexaploid wheat

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The long arm of chromosome 5A of common wheat carries the gene *Q* which is responsible for the suppression of speltoid effect and for squarehead spike characteristic of the variety Chinese Spring (SEARS 1954). In the present study, an attempt was made to map the genetic distance between the *Q* locus and centromere by using the telocentric method (SEARS 1966).

### Materials and Methods

Chinese Spring wheat ditelosomic for chromosome arm 5AL was crossed to the substitution line '*spelta*-5A'. Foundation seed of these two lines was kindly supplied by Dr. E.R. Sears, University of Missouri, Columbia, Mo., U.S.A. Spikes of the  $F_1$  plants ( $2n=41+t$ ) were speltoid and nonsquarehead. In the variety Chinese Spring, *Q* behaves as a partially dominant gene for the suppression of speltoid effect and as far as the squarehead effect is concerned, the gene is hemizygous ineffective and fully recessive. The  $F_1$  plants were selfed as well as test-crossed as male parents to Chinese Spring.

### Results and Discussion

The test-cross progeny consisted of 44 plants of which 26 were of *vulgare*-type and 18 of *spelta*-type. This phenotypic segregation conforms to a 1:1 ratio ( $X^2=1.46$ ;  $p=0.20-0.30$ ) indicating independence of the *Q* locus and the chromosome 5A centromere. The chromosome constitution and phenotype of the test cross plants are summarized in Table 1. The 16 *vulgare*-type plants with 42 normal chromosomes and the 2 speltoids with  $2n=41+t$  were scored as crossovers. The remaining 26 plants were non-crossovers. Out of the 32 plants with  $2n=42$ , the 16 *vulgare*-type plants were crossovers and the 16 *spelta*-type plants were non-crossovers. However, among the 12 monotelodisomic plants ( $2n=41+t$ ), only

Table 1. Chromosome numbers and phenotypes  
of test-cross progeny

Chromosome number	Phenotype	
	<i>Vulgare</i> -type (squarehead)	<i>Spelta</i> -type (non-squarehead)
$2n=42$	16	16
$2n=41+t$	10	2

Table 2. Chromosome constitution and phenotype of  $F_2$  plants

Chromosome number	Phenotype		$\chi^2$ (3:1)
	<i>Spelta</i> -type (non-squarehead)	<i>Vulgare</i> -type (squarehead)	
2n=42	20	7	0.01
2n=41+t	30	11	0.07
2n=40+2t	8	4	0.38
	58	22	0.27
$\chi^2$ heterogeneity=0.19; p=0.90-0.95			

the 2 *spelta*-types were crossovers. The reduced frequency of crossover telocentrics cannot be explained with the present data. Male transmission of telocentric-5AL in competition with complete 5A was 12/44 or 27.3%.

The  $F_2$  progeny of the selfed monotelodisomic  $F_1$  consisted of 227 plants of which 166 were of *spelta*-type (nonsquarehead) and 61 were of *vulgare*-type (squarehead). This segregation was a very good fit for a 3:1 ratio ( $X^2=0.424$ ;  $p=0.50-0.70$ ). A random sample of 80  $F_2$  plants were used for meiotic studies to determine their chromosome constitution. The chromosome numbers and phenotypes of these  $F_2$  plants are given in Table 2. Within each of the three chromosomal classes and over all 80 plants, the distribution of *spelta*-type: *vulgare*-type conformed with 3:1 ratio. The data of both test cross and  $F_2$  progenies indicated that the *Q* locus is genetically independent of the chromosome 5A centromere. Hence it can be concluded that the gene *Q* is located 50 or more crossover units from the centromere, i.e. it has a distal location on the long arm of chromosome 5A.

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## Effect of different plasmons on some metric traits in bread wheat (*Triticum aestivum*, L.)

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The cytoplasms of various *Triticum* and *Aegilops* species have been successfully used to induce cyto-male sterility in bread wheat (*Triticum aestivum*, L.). This has led to the production of completely male sterile analogues of wheat which may be used as female parents in hybrid seed production. Besides male sterility, some side effects of these s-cytoplasms on wheat plants are often observed and these may be attributed to cytoplasmic genes. This study was undertaken to study such effects on some metric traits in male sterile analogues of common wheat having various cytoplasms.

### Materials and Methods

The material for this study comprised 10 spring wheat cultivars including Sonalika, Sharbatai Sonara, Lerma Rojo, Hira, Norteno, N.P. 809, Kalyan Sona, K. 68, C. 306 and C. 591, and three male sterile cytoplasm sources, *viz.* *Triticum timopheevi*, *Aegilops ovata* and *Ae. caudata*. The s-cytoplasm sources were the lines having *T. aestivum* genome in the alien cytoplasm. These were used as female parents and were crossed with all the accessions of bread wheat. Subsequently five backcross doses to the recurrent parent, bread wheat in the present case, led to the production of male sterile analogues of wheat with three different cytoplasmic background.

The effects of these s-cytoplasms were recorded on male sterile wheat analogues for five morphological attributes, *viz.*, days to flower, plant height in cm., number of internodes per plant, effective tillers per plant and spike length in cm. Observations were recorded on 30 competitive plants spaced 30 cm between and 15 cm within the rows. Spike length, plant height and number of internodes were measured on the main tiller only.

### Results and Discussion

The effect of different s-cytoplasms on the 10 bread wheat cultivars for five morphological traits has been presented in Table 1. It is evident that *Ae. ovata* and *Ae. caudata* cytoplasms, though confer great luxuriance in terms of increased plant height and profuse tillering, cause prolonged flower initiation and delayed maturity which is not a desirable attribute from argonomic view point.

A perusal of the results (Table 1) shows that *Ae. ovata* and *Ae. caudata* cytoplasms caused an increase in ear length and number of productive tillers per plant. However, the



Table 1. Effect of different male sterile cytoplasm on some quantitative traits on bread wheat cultivars

Bread wheat cultivar	S-Cytoplasm	Characters				
		Days to flower	Plant height (cm)	Nodes/plant	Effective tillers/plant	Spike length (cm)
Sonalika	Normal	71.8	104.1	4.2	13.7	11.90
	<i>T. timopheevi</i>	90.9	115.5	4.0	23.1	13.85
	<i>Ae. ovata</i>	138.5	113.9	6.4	42.9	14.22
	<i>Ae. caudata</i>	139.3	109.4	4.5	44.6	10.81
Sharbati Sonora	Normal	66.7	83.5	4.3	21.8	11.31
	<i>T. timopheevi</i>	85.8	103.7	4.0	28.1	12.38
	<i>Ae. ovata</i>	137.9	108.5	6.4	34.6	12.95
	<i>Ae. caudata</i>	135.0	102.6	5.0	55.0	10.70
Lerma Rojo	Normal	89.5	107.1	4.6	15.3	12.96
	<i>T. timopheevi</i>	91.5	127.5	5.0	43.9	14.36
	<i>Ae. ovata</i>	135.5	96.2	6.0	55.2	14.22
	<i>Ae. caudata</i>	143.0	106.5	5.0	53.0	12.20
Hira	Normal	68.5	74.1	4.1	19.0	11.60
	<i>T. timopheevi</i>	77.9	74.3	4.0	23.3	12.28
	<i>Ae. ovata</i>	136.0	90.2	6.0	32.0	13.10
	<i>Ae. caudata</i>	135.0	98.9	5.0	64.0	10.25
Norteno	Normal	86.5	90.7	4.7	18.8	12.37
	<i>T. timopheevi</i>	99.8	112.1	5.0	23.0	14.82
	<i>Ae. ovata</i>	135.3	103.9	6.0	60.6	15.66
	<i>Ae. caudata</i>	143.5	114.8	5.0	62.5	10.90
N.P. 809	Normal	102.7	157.7	5.4	27.6	12.62
	<i>T. timopheevi</i>	108.3	141.5	5.3	36.5	14.32
	<i>Ae. ovata</i>	—	—	—	—	—
	<i>Ae. caudata</i>	142.0	112.6	6.0	72.0	13.50
Kalyan Sona	Normal	90.8	95.8	4.9	18.2	14.18
	<i>T. timopheevi</i>	89.6	120.8	5.0	27.2	14.12
	<i>Ae. ovata</i>	139.3	100.9	6.0	63.6	12.66
	<i>Ae. caudata</i>	—	—	—	—	—
K. 68	Normal	92.8	108.9	4.6	17.2	11.47
	<i>T. timopheevi</i>	84.0	112.6	5.0	28.5	14.67
	<i>Ae. ovata</i>	136.2	119.2	6.3	53.0	15.50
	<i>Ae. caudata</i>	—	—	—	—	—
C. 306	Normal	97.7	120.8	5.0	16.8	11.21
	<i>T. timopheevi</i>	84.2	100.8	4.0	28.4	12.72
	<i>Ae. ovata</i>	137.0	111.6	6.6	41.8	12.77
	<i>Ae. caudata</i>	—	—	—	—	—
C. 591	Normal	99.3	125.4	5.1	22.7	9.05
	<i>T. timopheevi</i>	107.4	134.1	4.0	29.8	12.90
	<i>Ae. ovata</i>	137.5	118.2	5.6	42.5	13.96
	<i>Ae. caudata</i>	—	—	—	—	—

Normal=*T. aestivum* cytoplasm.

seeds of these sterile analogues when obtained by crossing with maintainer or restorer lines, were highly shirvelled owing to their late maturity. This may be due to a sudden rise in temperature and a proportional decrease in relative humidity soon before the harvest time in late March and early April which caused forced maturity in the late maturing types.

This not only caused the production of shrivelled seeds but also caused a drastic decrease in grain yield per plant. A comparative performance of the first five bread wheat cultivars and their male sterile analogues for days to flower, effective tillers per plant and spike length in cm may be seen in Fig. 1.

*Triticium timopheevi* cytoplasm had relatively less pronounced effects than the *Aegilops* cytoplasm on the characters measured. This cytoplasm also increased effective tillers per plant and spike length in some cultivars but had equal maturity period and more or less the same plant height as the normals. Thus in comparison with normal plants with *T. aestivum* cytoplasm, the male sterile analogues with *T. timopheevi* cytoplasm were agronomically superior and could give highly yields when crossed with a diverse but effective

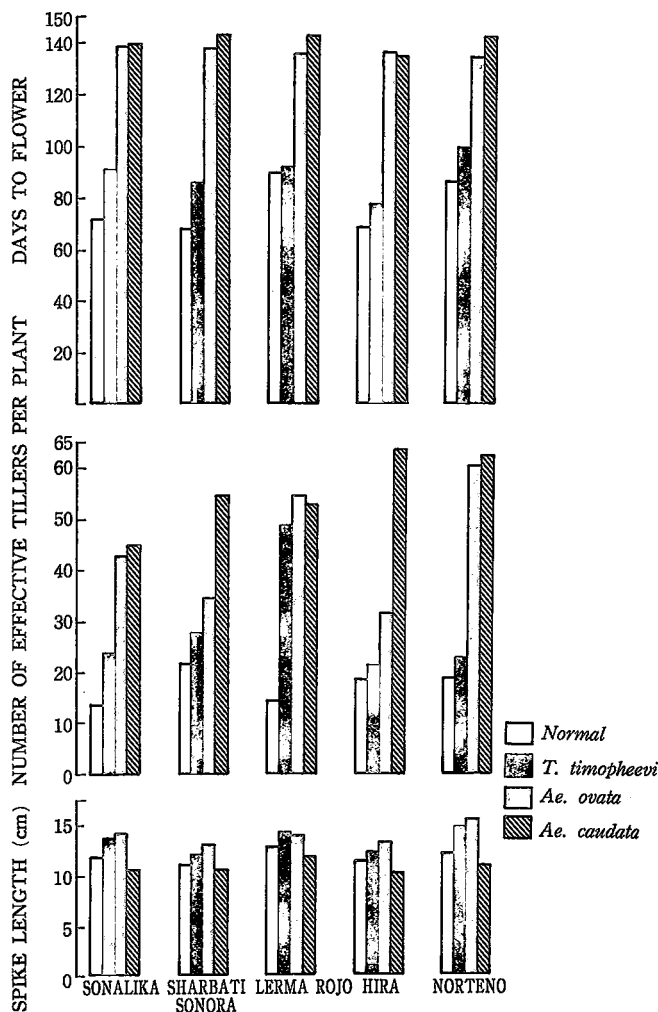


Fig. 1 Effects of three different s-cytoplasm on days to flower, effective tillers per plant and spike length in five wheat cultivars.

male fertility restorer line.

KIHARA (1966) first reported that various cytoplasms exerted different influences on substituted genomes. However, INGOLD (1968) observed that *Ae. ventricosa* cytoplasm did not cause major changes in the manifestation of substituted genomes. HORI & TSUNEWAKI (1969), TAHIR (1971) and MAAN (1979) studied the effects of s-cytoplasm and reported delayed maturity due to *Ae. ovata* cytoplasm. There appears to be a striking similarity between the results of this investigation and that of TAHIR (1971), that the cytoplasm of *Ae. ovata* delayed heading and increased tiller number and spike length in wheat. This may possibly be explained due to the common lineage between the materials studied in both the cases. He, however, did not study *Ae. caudata* cytoplasm which was found in the present study to transfer identical side effects as may be seen in Table 1.

Considering the side effects of male sterile cytoplasms of all the three species in the genomic background of bread wheat cultivars studied in this case it was concluded that *T. timopheevi* offered the best results of s-cytoplasm which carried plasmagenes that in interaction with the nuclear genes of *T. aestivum* improved many agronomic attributes besides conferring effective and stable male sterility in the bread wheat analogues.

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## Possibilities for increasing the content of lysine in the wheat grains

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The increase of protein content in the grain of the cereals, wheat inclusive, is an important problem in all breeding work. Breeders are searching for ways and methods to increase its quantity and to change its amino acid composition. Protein in the present wheat varieties contains smaller quantities of the irreplaceable aminoacids lysine, methionine, tryptophan, due to which its biological of full value reduces. Breeding for higher protein content is, however hampered by the polygenic character of its inheritance and by the significant protein phenotypic variation, dependent on the conditions of growing. A number of studies (ZAFGREN *et al.* 1968; LELATA *et al.* 1972) have proved the existence of negative correlation between productivity and protein content and between protein and amino acid composition of the grain.

Some results obtained by crossing mutants of high-protein content and different varieties show some advantages of the latter, when used as donors of high protein content and of certain protein components (SHOLZ 1976, ROBSON 1976)

Our task was, through the usage of mutagene factors, to develop wheat forms of higher lysine content and to transfer it to other varieties and lines.

On 1967 such a mutant of high lysine content M 1155 was developed by gamma irradiation of dry seeds of variety Skorospelka 35.

On 1969 the some mutant in  $M_2$  was selected, as a short-stemmed form. In 1972 the analysis of the biochemical qualities of a series of consolidated mutant lines in  $M_5$  established the high lysine content of M 1155. Irrespective of the variation of lysine in the separate years, it remains always over 3.2% of the protein, at 2.7% for the initial variety and 2.6% for the standard-Sadovo I. The newly developed high-lysine mutant line was however of some substantial defects, therefore it could not be used directly as a variety. So the same year it was included in hybridization together with other mutant lines of better economic qualities. A considerable number of progenis was selected in  $F_2$ , two of which at the preliminary analysis, manifested a more favourable combination of the desired characters.

That selection materials therefore got the numbers 61-779 and 64-779. Those number due to their morphological non-uniformity were divided into lines. Results for two successive years concerning the productivity and the protein and lysine content both in the initial forms and the lines studied are given in Table 1.

Both protein and lysine in the first combination were inherited intermediary. Their quantities were smaller than those of the high-lysine mutant but bigger than the same of the second component. Protein content in the second combination is lower than that of

both the parents, while respect to lysine content that combination is inferior to the high-lysine parent, but superior to the second parent. However, in all analysed lines of the two combinations, lysine content is over 3%.

A very favourable combination between protein and lysine exists in of the lines 61-779/401. The comparatively high protein content there corresponds to a high lysine content. That comes to show that hybridization with high-lysine wheat mutants may give, in some cases, transgressive forms of good productivity, high protein and lysine content and a favourable interrelation between protein and lysine in the grain.

Table 1. Results of the combination high-lysine mutant × mutants for two successive years average (1978-1979)

Parents and combinations	Grain yield (in c/I ha)	Protein in % to absolutely dry substance	Lysine	
			mg/100 g. abs. dry substance	% to protein
♂ M 1155	5585	15.85	555	3.51
♀ M 166	5530	13.61	337	2.48
61-779/401	6020	15.14	481	3.17
61-779/439	5680	14.76	486	3.27
♂ M 1155	5585	15.83	555	3.51
♀ M 567	5710	14.17	407	2.87
64-779/311	5480	12.82	400	3.11
64-779/327	6560	13.85	433	3.13

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## Inheritance of seed protein in winter and spring wheat crosses

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It is not a simple breeding goal to produce high yielding wheat lines with high protein content. The main limitations are the close negative correlation between yield and protein content (SHAHANI 1980), the polygenic inheritance of this trait (COWLEY & WELLS 1980; KERTESZ *et al.* 1980) and the influence of the environmental factors (CEAPOIU *et al.* 1975; KOUAME MIEZAN *et al.* 1977).

The present study was, therefore, designed to study the inheritance of protein content in wheat grain and to evaluate parents,  $F_1$  and  $F_2$  populations of two different spring and winter wheats in contrasting environments.

### Material and Methods

Two high yielding winter wheat lines (F310-C3-4 and F21-76), bred at Research Institute for Cereals and Industrial Crops, Fundulea, Romania were crossed direct and reciprocal with two semidwarf spring wheat varieties Pak-70 and Tandojam-75, bred at Research Institute, Tandojam, Pakistan. Seeds of  $F_0$  hybrids were sown immediately after harvest in phytotron. Parents,  $F_1$  and  $F_2$ 's were grown in October, 1978 and March, 1979 using randomized block design with three replications in order to study the biological material in two different contrasting environments. Protein content was determined by microkjeldahl method as crude nitrogen times 5.7. Data for grain protein percentage and protein per grain in milligrams were analyzed statistically. Estimates of broad sense heritability and genetic advances with selection intensity of 5% were computed as follows:

$$\text{Heritability \%} = \frac{S^2F_2 - S^2F_1}{S^2F_2} \times 100$$

where,  $S^2F_1$  and  $S^2F_2$  are the co-variances of  $F_1$  and  $F_2$  respectively. Genetic advance was calculated after LARIK *et al.* (1980) and computed by the following formula:

$$\text{G.A.} = (k) (Op) (H)$$

### Results and Discussion

#### Grain Protein Content Percentage

The results (Table 1) reveal that the spring wheat parents did not survive in autumn

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Table 1. Grain protein content in percentage of parents, F<sub>1</sub> and F<sub>2</sub> generations sown in autumn and spring seasons.

Parents	Autumn sowing				Spring sowing			
	Mean		S.D.		Mean		S.D.	
F310-C3-4	18.349		0.989		19.523		1.022	
F21-76	19.392		0.988		20.741		0.741	
Pak-70	—		—		15.809		0.802	
Tandojam-75	—		—		15.685		0.905	
Combinations	F <sub>1</sub>		F <sub>2</sub>		F <sub>1</sub>		F <sub>2</sub>	
	Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.
F310-C3-4 × Pak-70	17.003	0.956	16.373	1.415	16.676	0.980	16.731	1.606
Pak-70 × F310-C3-4	17.526	0.871	16.525	1.373	16.721	0.883	16.851	1.463
F310-C3-4 × Tandojam-75.	16.931	0.796	16.610	1.423	16.619	1.014	17.181	1.547
Tandojam-75 × F310-C3-4.	16.889	0.860	16.956	1.573	16.661	0.957	17.182	1.475
F21-76 × Pak-70.	17.162	0.988	17.455	1.662	16.662	0.939	17.139	1.507
Pak-70 × F21-76.	17.248	0.937	17.590	1.613	16.989	1.044	17.151	1.630
F21-76 × Tandojam-75.	16.967	0.884	16.957	1.402	17.079	0.927	17.427	1.579
Tandojam-75 × F21-76.	16.947	0.871	16.849	1.436	17.115	0.912	17.873	1.526

Table 2. Protein content per grain in milligrams of parents, F<sub>1</sub> and F<sub>2</sub> generations sown in autumn and spring seasons.

Parents	Autumn sowing				Spring sowing			
	Mean		S.D.		Mean		S.D.	
F310-C3-4	7.898		0.606		6.790		0.693	
F21-76	8.495		0.859		6.805		0.983	
Pak-70	—		—		6.363		0.830	
Tandojam-75	—		—		6.068		0.832	
Combinations	F <sub>1</sub>		F <sub>2</sub>		F <sub>1</sub>		F <sub>2</sub>	
	Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.
F310-C3-4 × Pak-70	6.825	0.732	5.456	1.163	6.567	0.706	5.779	0.854
Pak-70 × F310-C3-4.	6.755	0.712	5.765	1.121	6.200	0.872	5.831	0.969
F310-C3-4 × Tandojam-75.	6.642	0.769	6.404	1.111	6.541	0.931	6.051	1.069
Tandojam-75 × F310-C3-4.	6.139	0.725	5.936	1.049	6.495	0.985	6.529	1.156
F21-76 × Pak-70	7.337	0.980	6.354	1.671	7.363	0.991	6.241	1.183
Pak-70 × F21-76.	6.961	0.855	6.778	1.412	7.513	0.964	6.370	1.145
F21-76 × Tandojam-75.	7.578	0.876	6.600	1.051	7.239	0.893	6.152	1.121
Tandojam-75 × F21-76.	7.190	1.046	6.301	1.291	7.029	0.890	5.936	1.075

sowing. Increased grain protein percentage was noted in winter wheat parents, but line F21-76 was superior than line F310-C3-4 in both the seasons. The mean values of F<sub>1</sub> generations showed a partial dominance to the low grain protein content. The high values of protein percentage in spring sowing in comparison to winter sowing, are probably due to incomplete development of seeds caused by less period of standing crop. The differences between all the reciprocal crosses except F310-C3-4 × Pak-70 were small

Table 3. Estimates of heritability (b. s) and expected genetic advance with selection intensity (k)\* of five percent for grain protein percentage and protein per grain from various F<sub>2</sub> populations sown in autumn and spring seasons.

Sowing Time	Genetic estimates	F <sub>2</sub> Populations							
		F310-C3-4 × Pak-70	Pak-70 × F310-C3-4	F310-C3-4 × Tando-jam-75	Tando-jam-75 × F310-C3-4	F21-76 × Pak-70	Pak-70 × F21-76	F21-76 × Tando-jam-75	Tando-jam-75 × F21-76
<b>GRAIN PROTEIN PERCENTAGE</b>									
AUTUMN	Heritability % (b.s)	54.34	59.76	68.72	70.08	64.25	66.27	60.26	63.23
	Genetic advance.	1.584	1.690	2.014	2.271	2.700	2.202	1.740	1.870
SPRING	Heritability % (b.s)	62.75	63.55	55.07	57.90	61.21	59.01	65.50	64.23
	Genetic advance.	2.076	1.915	1.819	1.759	1.900	1.981	2.130	2.079
<b>PROTEIN PER GRAIN (milligrams)</b>									
AUTUMN	Heritability % (b.s)	60.33	59.66	52.13	52.22	65.56	63.35	30.61	34.21
	Genetic advance.	1.445	1.378	1.193	1.128	2.257	1.843	0.663	0.910
SPRING	Heritability % (b.s)	31.70	32.77	24.27	27.40	30.01	29.44	36.51	31.53
	Genetic advance.	0.558	0.654	0.534	0.652	0.731	0.696	0.843	0.698

and non-significant. This proves that there is no practical importance in reciprocal crossing for determination of grain protein content. It is concluded that two genetically different germplasms used in these crosses did not express at cytoplasmic level. The great difference among the mean values of hybrids of two different sowing times displayed that genotype × environment interactions played a major role in the expression of grain protein content. The environmental factors modify the behaviour of the parents and of the F<sub>1</sub> and F<sub>2</sub> generations. A great variation of F<sub>1</sub> populations in comparison to parents and F<sub>1</sub> populations, indicate the existence of a number of important genetic components which control the grain protein percentage in common wheat. Normal frequency distribution in F<sub>2</sub> populations suggests a complex genetic control of protein percentage in wheat.

The heritability and genetic advance values (Table 3) for this trait under two sets of environmental conditions are considerably high in F<sub>2</sub> populations. This suggests that F<sub>2</sub> populations contain a large genetical variability and one could expect a potential gain to be achieved through selection in F<sub>2</sub> populations. These results are in confirmation with JOHNSON *et al.* (1972) and SOOMRO & LARIK (1981).

#### Protein Content Per Grain

The data (Table 2) confirm that both winter lines (F310-C3-4 and F21-76) are superior



in absolute protein content than spring wheats. This proves their superior efficiency in nitrogen metabolism. The autumn sown winter parental lines had more protein per grain in comparison to spring sown parents. This is probably due to late maturity and incomplete development of grains in spring sowing. Protein per grain in autumn sown  $F_1$  hybrids does not reach even to the level of low grain protein content parent. Whereas, in spring sowing some of the crosses displayed partial dominance and the others heterotic effect for protein content per grain. Differences between the reciprocals of  $F_1$  and  $F_2$  populations for protein per grain are negligible and nonsignificant. This reflects that absolute protein content is not affected by the cytoplasm. All the combinations with a line F21-76 of  $F_1$  population, showed high mean values for absolute protein content. This indicates the genetic superiority of line F21-76 for protein per grain. The mean values of protein per grain in  $F_2$  generations of all the hybrids are low in comparison to  $F_1$  generations in both the seasons. This tendency of increase in protein percentage and decrease in protein per grain suggests that in segregated populations the small size and shrivalled seeds were developed.

Estimates of heritability and genetic advance in autumn sown  $F_2$  populations is considerably high as compared to spring sown population, indicating possible genetic diversity. The  $F_2$  populations of almost every cross approached to normal distribution, suggesting polygenic control of the trait. Since heritability and expected genetic advance for this trait in autumn sown  $F_2$  population is quite high as shown in Table 3, significant gain could be achieved through selection in future generations. On the contrary low heritability and low genetic advance in spring sown  $F_2$  populations indicate that the character could be transmitted to future generations, however, no significant gain could be achieved through selection in early generations. These results are in accordance with LARIK (1978) and SOOMRO & LARIK (1981).

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## Systematic spread of Karnal bunt (*Neovossia indica* (Mitra) Mundkur) disease of wheat

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Karnal bunt (*Neovossia indica* (Mitra) Mundkur) disease of wheat, identified as early as 1930 at Karnal in India, was mainly confined to northern India until 1960's. But now, it has spread to all the wheat growing states of India and a few other countries. The disease is not internally seed-borne. Its infection is reported to occur at the time of anthesis from the sickle-shaped primary sporidia produced from the soil borne chlamydo spores. Low temperature and high humidity at the time of anthesis favour maximum disease development (BEDI *et al.*, 1949). The infected grains are damaged to varying extent ranging from only a small black spot on the embryonic end to the entire grain being transformed into a bunt-ball. As a result of Karnal bunt disease the grain yield is reduced and the quality of wheat products is deteriorated.

In spite of its early identification and frequent epidemics in India, the mode of primary infection and further development of Karnal bunt has not been established on the firm footing. Infection has been reported to occur at the time of anthesis whereas maximum infection was obtained on artificial inoculation at the boot stage when there is no anthesis in the spike. It has been reported that the distribution of Karnal bunt infected grains in a spike is random and each floret gets infected by separate air borne sporidia at the time of its anthesis. The distribution of the Karnal bunt affected florets in the intact spikelets and spike under natural incidence has not been investigated so far.

The distribution of Karnal bunt infected florets (grains) under natural incidence of disease in intact spikelets and spikes was studied in wheat variety WL 711 (highly susceptible to Karnal bunt) in 1981 and 1982. In each year 10 spikes with naturally infected grains were selected from the seed multiplication plots. In each spike the row with the lower most fertile spikelet was designated as I and the opposite row as II. The data on number of spikelets per spike, number of infected/total grains per spikelet in each row from bottom to the tip of spike, number of primary infection sites and the percentage of spikelets infected per spike in 1981 and 1982 is given in Table 1 and 2 respectively. In each spike, the spikelets with maximum grain damage were assumed to be the primary infection sites (parenthesis).

The natural incidence of Karnal bunt was higher in 1981 than in 1982. Similarly the number of primary infection sites per spike in 1981 varied from 1-3 whereas in 1982 all the spikes analysed had only one primary infection site. In both years upper half of spikes had higher incidence of Karnal bunt than the lower half. The primary infection sites also

appeared to be slightly more frequent in the upper half than in the lower half. The primary infection appears to be more or less random as it can occur any where in the spike. Its higher frequency in the upper half of spike may be attributed to the higher number of florets and early anthesis in the upper half of spike. The upper half is also probably more exposed to air borne sporidia than the lower half. It is, however, not established at what stage the primary infection occurs as it has also been possible to get very high incidence of the disease on artificial inoculation at the boot stage (Aujla, pers. comm.).

The most interesting finding from the distribution of the infected florets in the intact spikelets and spikes is that the infected florets and spikelets are clustered around the spikelets of primary infection in each row and direction and are not distributed randomly as reported earlier. It was also found that the extent of damage to the seed from Karnal bunt reduced progressively as the distance of the infected florets increased from the primary infection sites. The spikelets at the primary infection site had mostly 100% of the florets infected while the infected spikelets away from the primary infection sites had some healthy grains. *This indicates that the Karnal bunt disease spreads systematically* to the adjacent florets and spikelets in each row in both directions of the spike from the primary infection site. Under the favourable environmental conditions the disease would spread to more spikelets and cause greater damage to infected grains. In 1981 when the environmental factors were more favourable, not only the spikes had higher number of primary infection sites but also had greater spread of the disease to the adjacent spikelets than in 1982.

There are 1-4 florets in a spikelet. It takes about 7 to 10 days for the completion of anthesis within a spikelet as it first starts in the floret nearest to the rachis and then in the 2nd, 3rd and 4th florets in each spikelet. The fact that 100% of the florets in the spikelets at primary infection sites and some adjacent spikelets are infected irrespective of 7 to 10 days difference in their anthesis, the probability of infection of individual florets in a given spikelet, with all infected florets, by separate sporidia at the time of anthesis is very very small as compared to the observed frequency, confirming that the spread of disease after primary infection is highly systematic.

The infected florets (grains) away from the primary infection site at different stages of grain development, such as dough had only a small black spot at the embryonic end indicating that the infection had just started. This shows that the secondary infection can probably take place at any stage of caryopsis development after anthesis.

From the observations reported here it can be concluded that Karnal bunt spreads very systematically to different florets and spikelets from the primary infection site (floret) and the secondary infection can take place at any stage of carropsis development after anthesis up to dough stage.

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Table 1. Distribution and extent of Karnal bunt infection in the spikelets at primary infection variety WL 711 in Rabi 1980-81. The extent of further spread of Karanal bunt

Spike No.	No. of spikelets per spike	Spike row	No. of Karnal bunt infected grains/total grains per spikelet.					
			1	2	3	4	5	
1	13	I	—	—	—	—	2/2	(3/3)
		II	—	—	—	—	—	2/2
2	17	I	—	—	—	—	—	—
		II	—	—	—	—	—	—
3	21	I	—	(4/4)	1/3	—	—	—
		II	—	1/2	—	—	—	—
4	18	I	—	—	—	—	—	—
		II	—	—	—	—	—	2/3
5	18	I	—	—	(3/3)	—	2/3	3/3
		II	—	—	—	—	(2/3)	3/3
6	19	I	—	—	—	—	—	—
		II	—	—	—	—	—	—
7	20	I	—	—	—	—	1/3	—
		II	—	2/4	4/4	—	1/4	—
8	16	I	—	—	—	—	—	(3/3)
		II	—	—	—	—	1/4	2/2
9	16	I	—	—	—	—	(4/4)	—
		II	—	—	—	2/3	2/3	—
10	17	I	1/2	(4/4)	—	—	—	—
		II	—	3/3	—	—	—	—

—: indicates the spikelets with

Table 2. Distribution and extent of Karnal bunt infection in the spikelets at primary infection in rabi 1981-82. The extent of further spread of Karnal bunt from the primary infec-

Spike No.	No. of spikelet per spike	Spike row	No. of karnal bunt infected grains/total grains per spikelet.									
			1	2	3	4	5	6				
1	18	I	0/3	0/4	0/4	0/4	0/4	0/3	0/2	0/2	0/2	
		II	0/3	0/4	0/4	0/4	0/4	0/3	0/2	0/2	0/2	
2	23	I	0/3	0/4	0/4	0/4	0/4	0/4	0/4	0/4	0/3	
		II	0/4	0/4	0/4	0/4	0/4	0/4	0/4	0/3	0/3	
3	18	I	0/2	0/4	0/4	0/4	0/4	0/4	0/4	0/4	0/4	
		II	0/4	0/4	0/4	0/4	0/4	0/4	0/4	(4/4)	0/4	
4	20	I	0/2	0/4	0/4	0/3	0/3	0/3	0/3	0/3	0/3	
		II	0/3	0/4	0/3	0/4	0/3	0/3	0/3	0/2	0/2	
5	15	I	0/2	0/2	0/2	0/2	0/2	0/2	0/2	2/2	2/2	
		II	0/2	0/2	0/2	0/2	0/2	0/2	0/2	(2/2)	0/2	
6	21	I	0/2	0/3	0/3	2/3	(4/4)	1/3	0/3	0/3	0/3	
		II	0/3	0/3	0/4	3/3	2/3	0/3	0/3	0/3	0/3	
7	20	I	0/2	0/4	0/4	0/4	0/4	0/4	0/4	0/4	0/4	
		II	0/3	0/4	0/4	0/4	0/3	0/3	0/3	0/4	0/4	
8	19	I	0/3	0/3	0/4	0/4	0/3	2/3	2/3	2/3	1/3	
		II	0/3	0/4	0/4	2/3	(3/3)	0/3	0/3	0/3	0/3	
9	16	I	0/3	0/3	0/4	0/3	0/3	0/3	0/3	0/3	0/2	
		II	0/3	0/4	0/3	0/3	0/3	0/3	0/3	0/3	0/2	
10	19	I	0/3	0/3	0/3	0/3	0/3	0/3	0/3	0/3	0/3	
		II	0/3	0/3	0/3	0/3	0/3	0/3	0/3	0/3	0/3	

tion site (within parenthesis) and adjacent spikelets in both rows of 10 spikes of wheat from the primary infection site is underlined.

Spikelets are numbered from 1-12 from the base to the tip of spike.						No. of primary infection sites/spike	Spikelets infected per spike (%)
6	7	8	9	10	11		
<u>2/2</u>	—					1	38.5
—	<u>2/2</u>	(3/3)	1/2	2/2		1	35.3
—	<u>3/4</u>	(3/3)	1/3			2	33.3
<u>3/3</u>	1/3	—	—	1/2	1/1	2	38.9
2/2	<u>3/3</u>	2/2	1/1	2/2	—	2	61.1
3/3	—	<u>4/4</u>	3/3	3/3	2/2	1	31.6
—	—	(4/4)	—	(3/3)	—	3	30.6
—	—	—	—	—	—	1	18.8
—	1/2	<u>2/2</u>	1/1	—	—	2	43.8
—	—	—	—	—	—	1	17.6

no Karnal bunt infection.

site (within parenthesis) and adjacent spikelets in both rows of 10 spikes of wheat variety WL 711 tion site is underlined.

Spikelets are numbered from 1-12 from the base to the tip of spike.						No. of primary infection site/spike	Spikelets infected/spike (%)
7	8	9	10	11	12		
<u>2/2</u>	<u>2/2</u>	1/2				1	22.2
0/3	0/2	<u>2/3</u>	2/3	0/2	0/2	1	17.4
0/4	0/3	0/2	0/3	0/2	2/2	1	5.3
0/2	0/3	<u>3/3</u>	0/2	0/2	0/1	1	5.0
1/1	1/1	0/1				1	26.6
0/3	0/3	0/2	0/2	0/2	0/2	1	23.5
0/4	1/3	<u>2/3</u>	2/3	0/2	0/2	1	25.0
0/3	0/2	0/3	0/2	0/2		1	26.3
0/3	<u>2/2</u>	(2/2)	1/1			1	18.8
(3/3)	1/3	0/2	0/2	0/2		1	10.5

## Performance of wheat and triticale cultivars subjected to soil salinity and soil moisture stress conditions.

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Farming in arid regions is completely dependent on irrigation water which normally carries high contents of salts in contrast to rain water. Although some of the ions which contribute to soil salinity may be indigenous in the soil, many are brought to soils in the irrigation waters. Poor drainage and high evaporation eventually accelerate salt accumulation.

Varietal response to salinity can play a key role in maintaining farming in arid regions using low quality irrigation waters. In the Central Region of Saudi Arabia, most farming soils are highly calcareous and considered saline or subjected to high salinity upon irrigation from sources with high content of salts. Therefore, it was found important to study the tolerance of some selected wheat and triticale cultivars to stress conditions induced by high soil salinity and limited soil moisture content.

### Materials and Methods

The experiment was carried out in greenhouse at the college of Agriculture, King Saud University. A representative soils sample was collected from the University Experimental Farm. A routine analysis was carried out in the laboratory and the main soil characteristics are shown in Table 1. Accordingly, the soil was considered non-saline

Table 1. Characteristics of the soil sample used.

a. Mechanical soil separates:		
Sand		69%
Silt		23%
Clay		8%
b. Soil texture:		
Sandy loam		
c. Soil moisture characteristics:		
Saturation		27.0%
Field Capacity		13.3%
Wilting point		3.1%
Available moisture		10.2%
d. CaCO <sub>3</sub>		37.3%
e. EC (mmhos/cm)		3.7
f. pH		7.8

(EC 3.7 mmhos), highly calcareous (37% CaCO<sub>3</sub>) and sandy loam in texture.

Four levels of soil salinity were achieved at 3.7 (control), 6.0, 8.5 and 11.0 mmhos/cm. The salinity levels were attained by adding a solution containing NaCl, CaCl<sub>2</sub>, MgCl<sub>2</sub> in amounts equivalent to 3:2:1 ratio, respectively. The moisture content in each level was kept more or less constant at three levels: 100%, 50% and 20% of available water. These levels were achieved by weighing the pots daily and the loss of water was compensated by adding enough water to reach the required moisture level of the soil.

Three wheat (*Triticum aestivum*, L.) and one triticale (Triticosecale Wittmack) cultivars were used. These were: Florence aurore, Super X, Arz and triticale line Armadillo "S" × 308-3N'.

A germination test was carried out in the laboratory. Twenty germination dishes each contained 20 seeds were prepared from each cultivar. They were arranged in five groups, each of four replications. These groups were allowed to germinate under saline solutions of; distilled water, 3.7, 6.0, 8.5 and 11.0 mmhos/cm. The dishes were kept in the dark at room temperature. Number of germinating seeds were counted twice after five and nine days. Only normal seedlings were counted.

The four lines were seeded in pots on December 3, 1978. After germinations, the pots were thinned to five plants in each. The 12 treatments, four salinity and three moisture levels, were arranged in a factorial experiment within each cultivar (four replications). A total of 192 pots occurred in the experiment.

Each pot received a nutrient solution containing 160 ppm N, 20 ppm P and 20 ppm K. The solution was divided into equal portions, then added after 15 and 56 days from planting. Micronutrients were all added at once in a concentration similar to Hoagland solution. The pots remained in the greenhouse throughout the whole experiment.

Fifty-six days after planting, before anthesis, one of the four replicates was harvested for dry matter content. The remaining three replications were harvested at maturity and data were recorded on the following parameters; spikes per pot, spikelets per pot, grain number per pot, grain yield per pot (g), and 100-grain weight.

A routine analysis of variance was applied to all data and means were tested by Duncan's multiple range test.

### Results and Discussion

The germination test indicates significant differences among cultivars in germination percentage due to some unknown factors but not salinity (Table 2). These results imply that wheat and triticale seeds were tolerant to high salinity (EC 11 mmhos/cm) during germination. BHUMBLA & SINGH (1965) however, noted that germination percentage of wheat at EC 12 and 16 mmhos/cm declined by 25% and 80%, respectively.

Interactions between soil salinity and available soil water induced significant effects on dry matter content (Fig. 1), grain yield, grain number, and 100-grain weight (Tables 2 and 3). The stress conditions caused by high soil salinity and limited soil moisture progressively decreased the dry matter content of the wheat plant. In this case, the triticale line was the

Table 2. Summary of analysis of variance for different parameters

Source of variation	% germination	spikes per pot	spikelets per pot	grain yield per pot	grain number per pot	100-grain weight
Cultivars	**	**	**	**	**	N.S.
Treatments	—	N.S.	**	**	**	**
A. Water	—	**	**	**	**	N.S.
Salinity	N.S.	N.S.	**	**	**	**
A. Water × Salinity	—	N.S.	N.S.	*	**	*
Cultivars × Treatment	—	N.S.	N.S.	**	**	**
Cultivar × A. Water	—	—	—	**	**	N.S.
" × Salinity	—	—	—	*	**	**
" × Salinity × A. Water	—	—	—	N.S.	N.S.	**

\*,\*\* Significant at .05 and .01 levels, respectively. N.S. Not significant.

Table 3. Mean performance for different parameters measured

Entry	Germination %	Spikes per pot	Spikelets per pot	Grain yield per pot (g)	Grain number per pot	100- grain weight (g)
Cultivars:						
Arz	100a	4.92a	47.77ab	2.02ab	68.42a	2.89a
Florence	99a	4.92a	43.15b	2.05a	63.64a	3.06a
Super X	90c	4.64a	45.29ab	1.85b	69.64a	2.52a
Triticale	95b	3.33b	35.63ac	0.53c	13.61b	2.90a
Treatments:						
Available water						
100% (Control)	—	4.75a	51.06a	2.19a	67.29a	2.99a
50%	—	4.39b	43.59b	1.59b	56.81b	2.82a
20%	—	4.21b	34.23c	0.97c	37.38c	2.72a
Salinity (mmhos)						
D.W.	95.0(a)					
3.7 (Control)	96.0a	4.52a	53.43a	2.07a	66.22a	3.40a
6.0	96.0a	4.47a	48.68a	2.17a	67.28a	3.29a
8.0	97.0a	4.47a	39.02b	1.39b	47.63b	2.88b
11.0	95.3a	4.33a	31.22c	0.67c	34.17c	1.81c

(1) D.W.: Distilled water.

(2) Means carryig different letters differ significantly at .05.

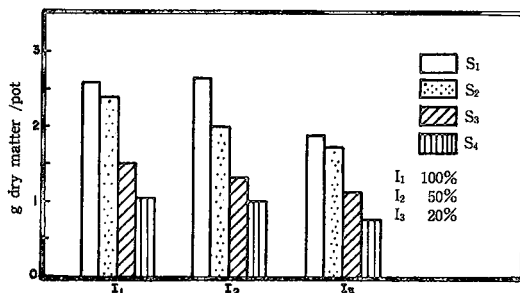


Fig. 1. Dry matter as affected by interaction of available weter (I) and salinity(S).



most tolerant to the highest salinity level though Super-X was more tolerant up to EC 8.5 mmhos/cm (Fig. 2).

Interactions of cultivar  $\times$  soil moisture and cultivar  $\times$  soil salinity were significant for grain yield, grain number and 100-grain weight (Fig. 3). This indicated that the response of these parameters to stress conditions were cultivar dependent.

On the basis of grain yield, the Florence line was found to be the most productive cultivar under stress condition caused by limited moisture and increased soil salinity followed by Arz and Super X, Table 3. When the relative yields (% of the control) were considered,

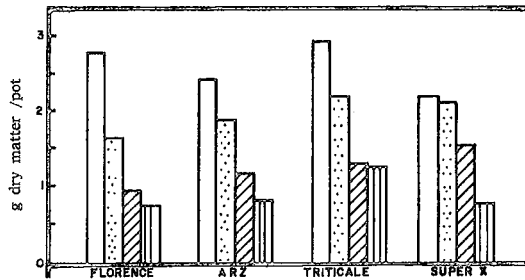


Fig. 2. Dry matter of different cultivars as affected by salinity.

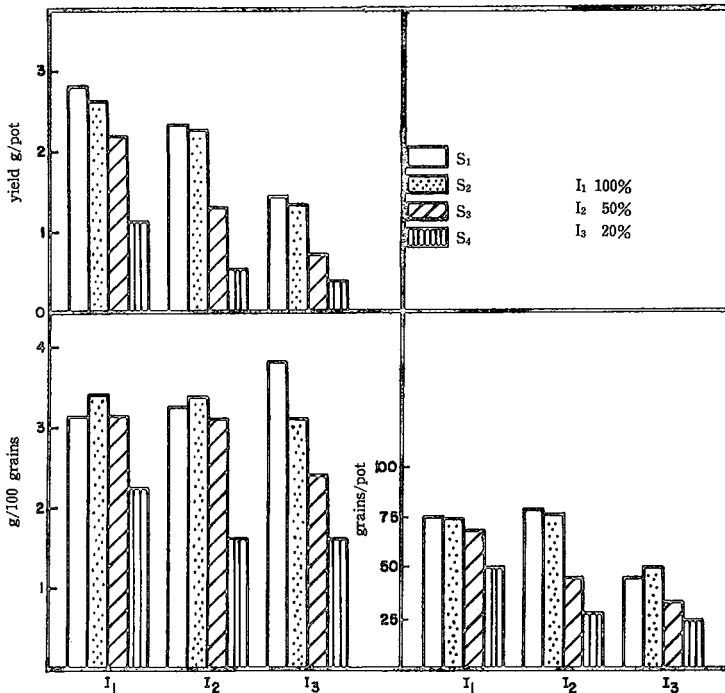


Fig. 3. Interaction of available water and salinity for grain yield, grain number and 100-grain weight.

the triticale line was found to be the most tolerant cultivar to stress conditions (yielding 62% of control), followed by Arz (59%), Florence (56%) and Super X (55%) (Table 4). The triticale line, however, showed a marked reduction in number of spikes and number of spikelets but less reduction in 100-grain weight compared to other cultivars which remained unchanged in the number of spikes. The differential response of grain yield and related characters in relation to soil stress conditions underline the genetic variability among the wheat and triticale cultivars thus warrant selection.

Reduction in soil moisture from 100% to 20% of the available water caused a remarkable loss in grain yield mounted to 56% of the control (Table 4). The number of spikes was reduced upon the decrease of available water from 100% to 50% only while spikelets and grain numbers were affected with each decrease in soil available water. Since the grain test weight remained independent from the change in soil available water, the loss in grain yield could be attributed to cumulative effects caused by reduction in spikes, spikelets and grain numbers, respectively.

Table 4. Performance of the four cultivars and characteristics (Percent of Control) as affected by treatments

Entry	Spike per pot	Spikelets per pot	Grain yield per pot	Grain number per pot	100-Grain weight
Cultivars:					
Arz	98. a	78. a	59. b	66. b	88. a
Florence	98. a	79. a	56. b	71. a	75. b
Super-X	99. a	81. a	55. b	73. a	72. b
Triticale	74. b	56. b	62. a	60. b	89. a
Treatments:					
Available water					
100% (control)	100. a	100. a	100. a	100. a	100. a
50%	92. b	85. b	73. b	84. b	94. a
20%	89. b	67. c	44. c	58. c	91. a
Salinity (mmhos/cm)					
3.7 (control)	100. a	100. a	100. a	100. a	100. a
6.0	99. a	91. a	105. a	102. a	97. a
8.5	99. a	73. b	67. b	72. b	85. b
11.0	96. a	58. c	32. c	52. c	53. c

\* Control values for each cultivar was obtained by averaging means of treatments EC 3.7 and 6. mmhos/cm at 100% available water level.

High soil salinity levels caused depressing effects on most characters (Tables 2, 3). Grain yield, number of spikelets, number of grains and test weight were all significantly reduced at salinity levels of EC 8.5 and 11.0 mmhos/cm. Thus, reduction in grain yield mounted to 33% and 68%, respectively (Table 4). It was noted that the reduction in grain yield resulted from marked reduction in number of spikelets 27% and 42% and number of grains 28% and 44% in the above mentioned salinity levels, respectively. Moreover, 100-grain weight was depressed by 15% and 47%, respectively. The depressing effects on 100-grain weight was attributed to the hastening of senescence (ASANA & KALE 1965; SARIN

& NARAYAN 1968) thus reducing the photosynthetic activity during grain development on the plants. It is noteworthy to mention that the number of spikes acted independently from the increasing levels of soil salinity. Another observation was that soil salinity level of EC 6.0 mmhos/cm was not different in its effects when compared with the control (3.7 mmhos/cm). In other words, all agronomic characters of the four cultivars were tolerant to soil salinity conditions upto 6.0 mmhos/cm without any detectable change. These results agree with previous reports (BERNSTEIN 1964; MAAS & HOFFMAN 1977) that salinity level of EC 6.0 mmhos/cm is the threshold at which yield of wheat will decrease upon any increase in salinity beyond the threshold. The rate of decrease in yield per mmhos/cm in the present study, however, was much higher (13.6%) than the 7.1% and 5.5% previously reported by the same authors.

The total soil water stress is determined by the algebraic sum of soil water tension plus the osmotic pressure of soil solution (BERNSTEIN 1974). As the salinity increases in the soil solution, available moisture in the soil should be increased to lessen the osmotic pressure of the soil solution so that water should be rendered more available for plant use. Under conditions similar to the study, soil moisture should not be reduced to less than 50% of the available water in order to avoid the effects of stress potential due to high salinity levels in the soil solution. It can be concluded that under high salinity conditions: (a) more frequent irrigation should be practiced, (b) cultivars with high salinity tolerance should be used.

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**Effectiveness of newly described leaf rust resistance genes against  
Indian cultures of standard races and biotypes  
of leaf rust in wheat.**

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Success in developing rust resistant cultivars depends upon a knowledge of effectiveness of the resistance genes against the local strains of pathogen in a geographic region. SAWHNEY *et al.* (1977) published a comprehensive account of seedling infections and field reactions of a series of isogenic lines/cultivars, each with a known leaf rust resistance gene, to Indian races of leaf rust pathogen. Lines carrying *Lr9* and *Lr19* were reported to be fully effective to all the 14 races/biotypes of leaf rust when tested in seedling individually and at adult plant stage in field conditions. Other isogenic lines/stocks except that with *Lr22* were reported to confer seedling resistance to one or more races of leaf rust. The designated series for leaf rust resistance genes has since been expanded. The information on all the presently known genes resistance to leaf rust for adult plant responses at several locations throughout the country was reported recently (SAWHNEY *et al.* 1982a). This communication reports on interactions of genotypes with the additional designation genes for resistance to leaf rust when tested in seedling individually with races/biotypes 12, 12A, 20, 77, 77A, 104, 104A, 107, 162, 162A, that are most prevalent and virulent in the country. The tests were conducted at temperature not exceeding 20°C. Seedling resistance is supplemented by adult plant response when tested in field conditions.

Table 1 lists the infection types produced on the genotypes each with known gene designated for resistance to leaf rust. It may be seen that a leaf rust resistance gene, *Lr24*, derived from *Agropyron elongatum*, and obtained in three stocks, Agent, Sear's 3Ag/3D, and white seeded recombinant (TR380-27×4/3 Ag 3-14) was observed fully effective to all the 10 races. Both Sear's 3Ag/3D and TR 380-27×4/3 Ag 3-14 were also reported to have complete resistance in seedling to all the currently maintained stem rust races in the country (SAWHNEY & GOEL 1981) and to both leaf and stem rust in adult plant stage (SAWHNEY *et al.* 1982 a and b). The stem rust resistance of these stocks is attributed to a known stem rust resistance gene *Sr24*. Both of these genes inherited together as a part of the alien chromosome sector (McINTOSH 1976). Use of white seeded stock (TR380-27×4/3 Ag 3-14) for breeding resistant varieties is of added advantage when white seeded varieties are consumers' preference.

*Lr25*: This gene was also observed to be completely effective when tested in 'Transac', a wheat line produced with a translocation of wheat chromosome 4A from non-homoeologous

Table 1. Infection types produced on stocks with known specific genes when tested with prevalent and virulent Indian cultures of standard races of leaf rust.

Stock	Races									
	12	12A	20	77	77A	104	104A	107	162	162A
'Agent' ( <i>Lr</i> 24)	0;-2	0;	0;	0;-1	0;-1	0;	0;	0;-1	0;	0;-1
Sear's 3Ag/3D ( <i>Lr</i> 24)	0;-1	0;-2	0;-2	0;-1	0;-1	0;-1	0;	0;-1	0;-2	0;-2
TR380-27 × 4/3Ag3-14 ( <i>Lr</i> 24)	0;	0;	0;	0;-2	0;-2	0;	0;-1	0;	0;	0;
'Transac' ( <i>Lr</i> 25)	0;	0;	0;	0;	0;	0;	0;-2	0;-1	0;-1	0;-2
'Kavkaz' ( <i>Lr</i> 26+ <i>Lr</i> 3)	0;	0;	0;	0;	0;	0;	0;	0;	0;	0;
'Gatcher' ( <i>Lr</i> 27)	0;-2	0;	0;-2	0;-1	4	0;-2	0;-1	0;-1	4	4
CS2A/2M 4/2 ( <i>Lr</i> 28)	0;	0;	0;	0;	0;	0;-1	0;	0;	0;	0;
CS7D/Ag #11 ( <i>Lr</i> 29)	0;	0;	0;	0;-1	0;-1	0;-2	0;-2	0;	0;-2	0;-2
'Terensio' ( <i>Lr</i> 30)	3	3	3	3-4	3	3	3	3	3	3
Chinese Spring	4	4	3	3	4	4	4	4	3	4

2R chromosome of Rosen Rye. The genotype was observed to produce low co-efficient of infection in field conditions (SAWHNEY *et al.* 1982a).

Cultivar 'Kavkaz' with *Lr*26 from rye was found to produce IT (0;) against each of the races tested. Furthermore, 'Kavkaz' was observed completely free of leaf rust infection in adult plant for the two seasons all over the country. Two of the other stocks with *Lr*26 (WRT ID/1R and 'Benno'), however, were observed to produce infection of low intensity which suggests that 'Kavkaz' has possibly additional gene(s) for resistance to leaf rust. This additional resistance is most probably due to complementary or additive gene interaction with *Lr*3, another leaf rust resistance gene known to be present in 'Kavkaz'

Cultivar 'Gatcher' with *Lr*27 was observed to be ineffective to some of the prevalent races/biotypes of leaf rust (77A, 162, and 162A) in the country. The cultivar was, however, observed to produce a low level of infection to leaf rust in field tests (SAWHNEY *et al.* 1982a). This behaviour of 'Gatcher' for its resistance to leaf rust requires more detailed study.

Two stocks, CS 2A/2M 4/2 and CS 7D/Ag # 11, with *Lr*28 and *Lr*29 respectively were also found to be completely effective in seedling. Both of these genes were derived from *Aegilops speltoides* and present in these stocks in the background of Chinese Spring. In field tests, the low co-efficient of infection observed at different locations in the country further showed that both the stocks are also effective to leaf rust in adult plant.

Cultivar 'Terensio' with *Lr*30 was observed to produce compatible but low reactions against most of the races/biotypes tested in seedling. The line was also observed to produce moderate co-efficient of infection in field condition at adult plant stage. This gene appears to have relatively low value in breeding resistant cultivars in the country.

The identification of a number of potentially useful genes resistant to leaf rust and associated resistance to both leaf and stem rust reported in the present study should be of immense use in breeding rust resistant cultivars in the country.

### Acknowledgement

We thank Dr R.A. McINTOSH, University of Sydney for the supply of seed material and Dr S. NAGARAJAN, IARI Regional Station, Simla for the supply of initial inoculum of the rust pathogne.

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**Genetic analysis of resistance in some varieties of bread wheat  
(*Triticum aestivum*) to three races of yellow rust (*Puccinia striiformis*)\***

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In breeding programmes aimed at incorporating yellow rust resistance in wheat, genetic stocks are used that show resistance in the adult plant stage over a number of locations and seasons. In these stocks, little genetic information on Yr-genes is available even though information of this type is very essential to ensure genetic diversity in the programmes. The purpose of this study was to determine the inheritance of resistance to yellow rust in bread wheat.

#### Material and Methods

Seven bread wheat varieties, viz. CPAN 1360, CPAN 1444, Sonalika, HD 2009, Tanori 71, Tobari 66 and Kalyansona with undetermined Yr-genes, five varieties, viz. Bon Fermier (yr3a), Opal (yr4b), Lee (yr7), Compair (yr8) and Nudif TP 250 (Yr1+Yr6) and Agra Local, the universal susceptible variety, comprised the material for the present study. Seedling reactions were recorded on parental lines F<sub>1</sub> hybrid and F<sub>2</sub> progeny of several crosses. The details of the setting up of the laboratory and the recording of the observations are already on record (KOCHHAR, GILL & NANDA 1982).

#### Results and Discussion

The varieties CPAN 1444, Sonalika and Tobari 66 probably possess Yr3a as no segregation was observed in their crosses with tester line Bon Fermier against any of the three races studied. Varieties HD 2009 and Tanori 71 may also be presumed to carry this gene as indicated by no segregation in F<sub>2</sub> of their crosses with Bon Fermier (Yr3a) against two of the three races. A 63:1 ratio was observed for HD 2009 × Bon Fermier against race 31 and for Tanori 71 × Bon Fermier against race 38A. These varieties are thus suggested to have other loci for resistance against the respective races. It is rather unusual that susceptible seedlings should have been scored even when both the parents have been postulated to carry Yr3a. It may be due to some unknown inhibitory genes operating selectively, or Yr3a may be a complex locus, or it may have been altered in its reaction due to the residual genetic background. Similar results have been reported on wheat (LUPTON & MACER 1962; JAIN & GANDHI 1978). On a similar assumption varieties CPAN 1444, Sonalika, HD 2009 and Tanori 66 may be postulated to carry Yr4b which determined the

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\* A part of the Ph. D. Thesis of the Senior Author submitted to the Punjab Agricultural University.

resistance in the crosses of these lines with the tester line Opal against one or more races. However, the parentage of these varieties did not indicate any common parents which could be postulated to have contributed Yr4b. A segregation for two loci was observed in the cross CPAN 1360 × Opal against race 38A indicating that two other loci operated in determining the resistance in the cross against this race.

Since all the three races are virulent on variety Lee (Yr7) the segregation for this locus cannot be detected. However, a segregation of one locus in the cross CPAN 1360 × Lee against race 38A; of two loci in the cross Tobar 66 × Lee against race 20A, in cross Sonalika × Lee against race 31; and in crosses CPAN 1444 × Lee and HD 2009 × Lee against race 38A, and three loci in the crosses; in CPAN × Lee (for races 20A and 31) and Tobar 66 × Lee (against race 20A) suggested the presence of resistance genes other than Yr7 in the corresponding parents.

In their crosses with Compair (Yr8), the varieties CPAN 1360, CPAN 1444, DH 2009 and Tenori 71 showed two to three gene segregation indicating that genes other than Yr8 occur in these varieties giving resistance to race 38A.

Table 1. Seedling reaction of varieties with undetermined genes for resistance, tester Yr-lines and F<sub>1</sub> hybrids between varieties and testers and Agra Local against races 20A, 31 and 38A of *Puccinia striiformis*

Varieties Races		Tester Yr-Lines						
		Parental reactions	Bon Fermier (Yr3a)	Opal (Yr4b)	Lee (Yr7)	Compair (Yr8)	Nudif TP 250 (Yr1 + Yr6)	Agra Local (+)
	20A		0	0	4	3	0	4
	31		0	0	4	4	0-2	4
	38A		0	0	3+-4	0	0	4
CPAN	20A	0	0	0	0	0	-	-
1360	31	0	2	0	1	2	-	-
	38A	0	0	0	2	0-2	-	-
CPAN	20A	0	-	0	0	-	-	0
1444	31	0	-	0	0-1	2	-	1
	38A	0	-	0	0	0	-	0-2
Sonalika	20A	0	0	0	0	-	-	-
	31	0	0	0	1-2	-	0	-
	38A	0	0	0	-	-	0	-
HD	20A	0	-	0	0	-	0	0
2009	31	0	-	0	0-1	1	-	0-2
	38A	0	-	0	0	0	0	0
Tanori	20A	0	0	-	0	0	0	0
71	31	0	0	0	0	1-2	0	0
	38A	0	0	0	0	0	0	0
Tobar	20A	0	0	0	0-2	0-2	-	-
66	31	0	0	0	0	1-2	0	1-2
	38A	0	0	0	0	0	-	0

0-4=reaction types. -- = reaction not recorded.



Table 2. Segregation behaviour of crosses between varieties with undetermined res-genes and tester lines when tested at seedling stage in  $F_2$  against three races of *P. striiformis*

Cross		Number of seedlings				Ratio	Pvalue
		Observed		Expected			
		Res.	Susc.	Res.	Susc.		
<u>Race 20A</u>							
CPAN × 1360	Opal	201	0	—	—	—	—
	Lee	196	3	195.9	3.1	63:1	.98-.95
	Compair	156	41	147.7	49.7	3:1	.20-.10
Sonalika × Compair		196	5	197.9	3.1	63:1	.30-.20
HD 2009 × Compair		193	3	192.9	3.1	63:1	.98-.95
Tanori 71 × Compair		194	10	191.2	12.8	15:1	.50-.30
Tobari 66 × Lee		156	11	156.6	10.4	15:1	.90-.80
<u>Race 31</u>							
CPAN × 1360	Lee	168	3	168.3	2.7	63:1	.90-.80
	Compair	175	23	176.4	21.6	57:7	.80-.70
CPAN × 1444	Lee	180	12	180.0	12.0	15:1	.10-.00
	Compair	169	17	174.4	11.6	15:1	.20-.10
Sonalika × Lee	Compair	181	17	185.6	12.4	15:1	.20-.10
		193	5	194.9	3.1	63:1	.30-.20
HD 2009 ×	Bon Fermier	189	2	188.0	3.0	63:1	.70-.50
	Lee	174	19	180.9	12.1	15:1	.05-.02
	Compair	138	51	141.7	47.3	3:1	.70-.50
	Nudif TL 250	197	2	195.9	3.1	63:1	.70-.50
Tanori 71 ×	Opal	176	22	176.4	21.6	57:7	.95-.90
	Compair	181	22	180.8	22.2	57:7	.98-.95
<u>Race 38A</u>							
CPAN × 1360	Opal	185	9	181.9	12.1	15:1	.80-.70
	Lee	143	50	144.8	48.2	3:1	.80-.70
	Compair	160	28	167.4	20.6	57:7	.10-.05
CPAN × 1444	Lee	194	10	191.3	12.7	15:2	.50-.30
	Compair	196	3	195.9	3.1	63:1	.95-.90
HD 2009 ×	Lee	185	17	189.4	12.6	15:1	.30-.20
	Compair	185	8	180.9	12.1	15:1	.30-.20
Tanori 71 × Bon	Fermier	198	5	199.8	3.2	63:1	.50-.30
	Compair	179	11	178.1	11.9	15:1	.80-.70
Tobari 66 × Lee	Nudif TL 250	196	4	196.9	3.1	63:1	.70-.50
		179	3	179.1	2.9	63:1	.45-.90

The Nudif TP 250 genes (Yr1+Yr6) are indicated to be present in varieties CPAN 1444, Sonalika, HD 2009, Tanori 71 and Tobari 66 on the similar assumptions made above.

A limitation in the present set of materials may be attributed to the lack of information on the genetic nature of variety Agra Local, the susceptible variety used in the experiment. It was susceptible to the races used in the present study but this cannot be taken as an evidence that the variety is universal susceptible. Moreover, Lee (Yr7) was also susceptible to the three races. Studies of SAWHNEY & LUTHRA (1970) have already hinted at the

resemblance of reaction pattern of Agra Local with that of an International differential variety for yellow rust (Michigan Amber). The latter variety is suggested to carry Yr2+Yr7 (Anonymous, 1979). So long as a parasite culture carrying a matching virulent gene is not used, the capacity of the resistance gene in Agra Local, if present, to confer resistance will remain undiscovered and the particular allele will segregate as recessive alleles for susceptibility. This point gains favour from PERSON & MAYO's (1974) explanation on the genetic limitations on models for specific interactions.

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## II. Record

### Proceedings of the 17th Wheat Genetics Symposium of Japan

The 17th Wheat Genetics Symposium of Japan was held at Faculty of Agriculture, Kyoto University on Oct 9 and 10, 1982. The followings are the abstracts of the invited and contributed presentations. In addition to these contribution, slide demonstrations were presented by Dr. I. Nishiyama entitling 'Genetical utilization of the interspecific cross incompatibility', and by Drs S. Ohta and S. Sakamoto entitling 'Botanical expedition to Greece'.

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### Genetical analyses of dwarfism in common wheat

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Characterization, mode of gene action, and gene localization of a dwarf wheat variety, Aibian-I were investigated, which was obtained from Shei-Bei Agricultural Academy (San-Shi, China).

Aibian-I is a weak winter type wheat showing 32 cm height on average with normal size (7.2 cm on average) of spike on the reduced length of internodes. The number of node is not different from those of other varieties. The number of spikelets per spike is, also, comparable to other conventional varieties. It flowers without complete head emergence, but bears almost full set of seeds.

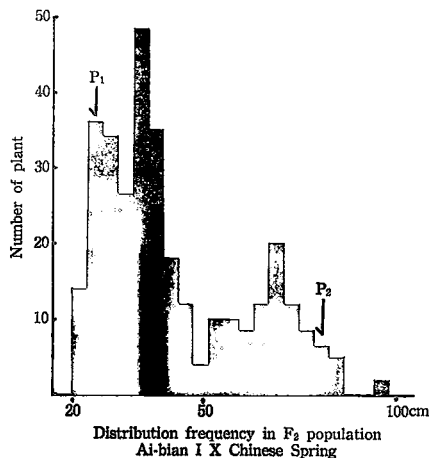


Fig. 1

Numerical analysis was conducted in 40 common wheat lines, including Aibian-I and 29 semi-dwarf varieties which were obtained from Japan, England, Holland, West Germany, USA, and Canada, on 25 quantitative characters. A dendrogram was drawn which classified them into four groups. The first group consists of 10 lines including four Canadian winter wheat varieties and experimental lines of Chinese Spring, Tve, *macha* and *spelta*. The second one includes most of semidwarf conventional varieties. Dwarf Bezostaya (a dwarf strain originally from USSR) was included into this group together with conventional varieties like Nugaines (USA), Hobbit (England), and Horoshiri (Japan). The third one is characteristic to include breeding parental lines of semi-dwarfness like Daruma, and Norin-10. Sonora-64, Norin-61, and Feilder were combined with these lines. The fourth consisted of only one line of Aibian-I, which was combined with other groups at most distance in the dendrogram. This result suggests that Aibian-I has different genetic components from others, and that there are some genetical differentiation of plant structure between the most of conventional varieties and their breeding parents.

F<sub>2</sub> segregation and monosomic analysis of Aibian-I showed that its dwarfness was controlled by a complete dominant single gene located on chromosome 4D. This fact also indicates that this dwarf gene is completely different from those of Norin-10, although one of them are located on 4D (GALE 1975). A dwarf wheat Tom Thumb is reported to have an incomplete dominant gene on chromosome 4A (MORRIS *et al.* 1975). Since nine induced dwarf or semi-dwarf genes were so far identified including induced mutations (reviewed by KONZAK 1975, 1980), the dwarf gene of Aibian-I could be designated as *Rht*<sub>10</sub> located on chromosome 4D (probably 4D<sup>s</sup> by a preliminary examination of telocentric mapping).

Sensitivity test of gibberellic acid was applied to seedlings of Aibian-I, which indicated that it was insensitive to GA<sub>3</sub>, and that the sensitivity segregated in the same fashion as dwarfness in F<sub>2</sub> generation.

Since Aibian-I has potential characters for wheat breeding like complete fertility, strong staw and stable dwarfness, the isogenic line and chromosome substitution line in Chinese Spring background are now being established as well as the introduction of the dwarf gene into the conventional varieties. Plants of these backcrosses at B<sub>3</sub> generation had 40-50 cm height with completely emerged head on strong straw having complete fertility.

### Progeny of a haploid common wheat with *Aegilops kotschy* cytoplasm

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The cytoplasm of *Aegilops kotschy* was introduced into a 1B/1R translocation line of common wheat, *Triticum aestivum* cv. Aurora by repeated backcrosses. In B<sub>3</sub> generation,

Table 1. Results of the cross, haploid of (*kotschyi*)-Aurora  
× normal Aurora

Category	Chromosome number	No.	%	
Total pollinated		1708	100.00	
Without seed set		1618	94.73	
With seed set		90	5.27	
Germless seed		19	1.11	
Seed not germinating		8	0.47	
Seed germinated		63	3.69	
Diploid	{ 44	1	} 30	1.76
	{ 43	1		
	{ 42	20		
	{ 41	6		
	{ 40	2		
Haploid	{ 22	2	} 28	1.64
	{ 21	23		
	{ 20	3		
Twin	{ 21, 42	2	} 5	0.29
	{ 21, 2n	1		
	{ 20, 41	1		
	{ ?, ?	1		

Table 2. Proportion of the different meiotic products at tetrad stage in the haploid of (*kotschyi*)-Aurora

	Monad	Dyad	Triad	Tetrad	Pentad	Hexad	Total
No.	1	15	30	926	53	7	1032
%	0.1	1.5	2.9	89.7	5.1	0.7	100.0

(*kotschyi*)-Aurora was the 1B/1R homozygote and produced haploids at a high frequency (93.8%). The resulting haploids were backcrossed with normal Aurora pollen. Results of the cross are shown in Table 1. Of 1708 florets pollinated, only 90 set seeds (5.3%). Among them, 19 were germless grains. 71 seeds were sown, of which 63 germinated. The chromosome numbers of plants were determined from root-tip mitosis. 30 were diploid (chromosome number=40-44) and 28 were haploid (20-22). Of five twin pairs, four were n-2n type.

From the crossed results, the egg cells with chromosome numbers ranging from 19 to 23 could take part in the fertilization. The observed frequency of female gametes with 21 chromosomes was extremely high in comparison with the theoretical one estimated from the binomial distribution. This is mainly due to the production of unreduced gametes following failure of the first or second division during meiosis. And also, the origin of aneuploid gametes (n=20 or 22) may be attributed to abnormal behavior of a univalent in meiotic division as proposed by SEARS (1939).

The haploid of (*kotschyi*)-Aurora had 21 unpaired chromosomes at metaphase I in 54.2% PMCs, while other had one or two bivalents. Metaphase cells with 21 univalents were

of two types: one showed normal univalents, and the other showed univalents with visible chromatids like those at mitotic metaphase. In the latter cells, these univalents divided equationally at the first division, and then restitution nuclei were formed at a low frequency because of an omitted second meiotic division. In fact, at tetrad stage, the frequency of dyads which were thought to be the product of unreduced gametes in microsporogenesis of the haploid was 1.5% (Table 2).

Non-pollinated florets of the haploid of (*kotschyi*)-Aurora were fixed in Carnoy's solution eight days after emasculation. Development of embryo in non-pollinated ovules of the haploid was studied anatomically using ordinary paraffin sectioning method. Of 129 ovules examined, 102 had no embryosac. Parthenogenetic development of embryo was found in five ovules (3.9%). In the remaining 22 ovules, no egg cell was developed. This fact indicates that the haploid formation in the haploid of (*kotschyi*)-Aurora is due to parthenogenesis of the egg cell similar to that found in the diploid of (*kotschyi*)-Aurora.

The authors wish to thank Dr. I. PANAYOTOV, Institute for Wheat and Sunflower, Bulgaria for supplying seeds of the 1B/1R heterozygous (*kotschyi*)-Aurora. This study was supported in part by a Grant-in-Aid (No. 57760006) from the Ministry of Education, Science and Culture, Japan.

### Cytoplasmic mutation induced by chemical mutagens

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It is known that cytoplasmic gene or genes have an important role in the inheritance of some characters such as male sterility, disease resistance and chlorophyll aberration. In this paper, the author wish to report the induction of the cytoplasmic mutation related to the male sterility in sugar beets. These informations may be worthwhile for the breeding of hybrid wheat by the use of cytoplasmic male sterility.

Three kinds of chemical mutagens, acriflavine, streptomycin and ethidium bromide were applied to seeds by the following procedures. Seeds of the maintainer strains having *N* cytoplasm dipped in different concentrations of aqueous solutions of acriflavine and streptomycin for 24 hours at 30°C in a dark room. The ethidium bromide treatment at 2000 ppm was carried out in a dark room kept at 5°C for 40 hours. After the treatments, seeds were washed in running water for 30 minutes. Photohermal induction was applied for the acceleration of the generations.

Male sterile plants (S.S.b or C.S.) occurred in  $M_1$  generation which were raised from seeds treated with the chemical mutagens. The progenies of male sterile plants were examined under open pollination to detect the possibility of transmission to the next generation. All

Table 1. Inheritance of male sterility induced by chemical mutagens

a. From  $M_1$  plants to  $M_2$  lines

Chemical	$M_1$ plant*	Male sterile type in $M_2$				Total
		N	S.S.a	S.S.b	C.S.	
Acridlavine (1500 ppm)	TK 81-0:362	2	7	18	2	29
Streptomycin (1500 ppm)	TK 76-0:3056	1	3	6	0	10
"	TK 76-0:3089	3	2	8	0	13
"	TK 76-0:1690	0	3	15	1	19
"	TK 81-0:1725	0	3	8	13	24
Ethidium bromide (2000 ppm)	TK 81-0:2133	0	0	1	19	10

\* Showing S.S.b type.

b. Test crossings with type-0 plant ( $N rf_1rf_1rf_2rf_2$ )

$M_2$ plant*	N+S.S.a	S.S.b	C.S.	Total	Genotype
$M_2$ ; 362- 837	0	0	15	15	$S rf_1rf_1 rf_2rf_2$
$M_2$ ; 362- 869	0	0	76	76	do.
$M_2$ ; 362- 881	0	0	16	16	do.
$M_2$ ; 362- 903	0	0	106	106	do.
$M_2$ ; 1690-1090	0	1	97	98	do.
$M_2$ ; 2133- 2	0	0	46	46	do.
TK76 MS;7 (check)	0	0	43	43	do.

\* Showing C.S. type.

S.S.b plants from the check strains did not transmit the male sterility to the next generation, while six S.S.b or C.S. plants in  $M_1$  transmitted the male sterility to  $M_2$  lines. By using complete sterile plants segregated from  $M_2$  lines, test crossings were tried with type 0 plants which have the genotype,  $N rf_1rf_1rf_2rf_2$ . In the cross combinations between C.S. plants in  $M_2$  and type 0 plants, most of the progenies produced 100% of C.S. plants (Table 1). Therefore it is assumed that the genotype of C.S. plant from mutant lines is  $S rf_1rf_1rf_2rf_2$  as well as that of the CMS strain of spontaneous origins. Cytoplasmic inheritance was confirmed by the reciprocal crossings between the male fertile plants which were segregated from  $M_2$  lines and the type 0 plants with  $N$  cytoplasm. The segregation of male sterile type differed prominently between reciprocal crossings indicating maternal inheritance of the male sterility. Then the evidence remains consistent with that the cytoplasmic mutation from  $N$  to  $S$  cytoplasm was induced by chemicals.

Though the cytoplasmic alternation or changes on male sterility in sorghum (ERICHSEN & ROSS 1963, MALINOVSKY *et al.* 1973) and pearl millet (BURTON & HANNA 1976) are reported, the proof of cytoplasmic inheritance has remained insufficiently without confirmation. However, it was evident in this experiment that the chemical mutagens induced the cytoplasmic mutation on male sterility as well as gamma rays (Kinoshita and Takahashi 1969).

It is known that acridine dyes are potent inducers of the *petite* mutation in mitochondria

genome of yeast and that streptomycin acts as a highly effective mutagen for chloroplast genes in *Chlamydomonas* (SAGER 1972). In the present experiment, the mutagens were effective for induction of the mutation for the cytoplasmic gene in higher plants.

### Diversity of the chloroplast genome among *Triticum* and *Aegilops* specie revealed by chloroplast dna restriction fragment patterns

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The chloroplast genome of higher plants has homogeneous circular molecules ranging in size from 120 kbp in pea to 180 kbp in *Spirodela*. Chloroplast DNA (ctDNA) examined mostly contains a large inverted repeat sequence of 22–25 kbp, in which ribosomal RNA genes are involved. The chloroplast genome of wheat contains 135 kbp molecules and segment of the inverted repeat is 21.0 kbp in common with many other higher plant ctDNAs (BOWMAN *et al.* 1981). I present here the chloroplast genome diversity and phylogenetic relationships among *Triticum* and *Aegilops* species by comparing ctDNA restriction fragment patterns.

Intact chloroplasts were isolated from leaves of 34 self-fertile alloplasmic as well as from six euplasmic lines of wheats (almost all *Triticum* and *Aegilops* species), followed by the method of KOLODNER and TEWARI (1975). Chloroplasts were purified from crude preparations using a discontinuous gradient made with 10, 40 and 75% Percoll solutions. Purified chloroplasts were lysed in a solution containing sodium lauryl sarcosinate and Proteinase K. Then, ctDNAs were isolated. The restriction patterns of these ctDNAs digested with eight restriction endonucleases (*Bam* HI, *Eco* RI, *Hind* III, *Kpn* I, *Pst* I, *Sal* I, *Sam* I, *Xho* I) could be classified into 13 types, in total. The physical maps constructed by *Pst* I and *Sal* I of ctDNAs of common wheat, *Ae. squarrosa* and *Ae. uniaristata* were about 0.2–0.5 kbp smaller than that of most other species. These classification of ctDNAs was principally in agreement with that of cytoplasm which is based on nuclear-cytoplasm interactions. Most polyploids and their related diploids showed identical restriction patterns indicating the conservatism of chloroplast genome during speciation.

The restriction fragment patterns of Emmer and Dinkel (common) wheats were identical with those of *Ae. longissima*, and different from those of all other diploids. The restriction fragment patterns of Timpheevi wheats were identical with those of *Ae. aucheri*. *Ae. searsii* and *Ae. bicornis*, and *T. urartu* and *T. monococcum* had the identical chloroplast genomes with each other.



## Organization and expression of chloroplast genomes

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Recent progress in molecular biological analyses of chloroplast DNAs have revealed many interesting features of these genomes. Chloroplast DNAs of higher plants are double-stranded circular DNA with about 160,000 base-pairs (bp) length, and have a long inverted-repeat sequence (22,000 bp) (Fig. 1). Approximately 100–200 genes are thought to be encoded in chloroplast DNA. In this article, we refer to organization and expression of genes for the large subunit (LS) of ribulose-1,5-bisphosphate carboxylase/oxygenase, rRNA and tRNA of tobacco chloroplast.

We have cloned and sequenced the LS gene to tobacco chloroplast and its flanking regions. The coding region contains 1431 nucleotides (477 codons). Sequences of the LS

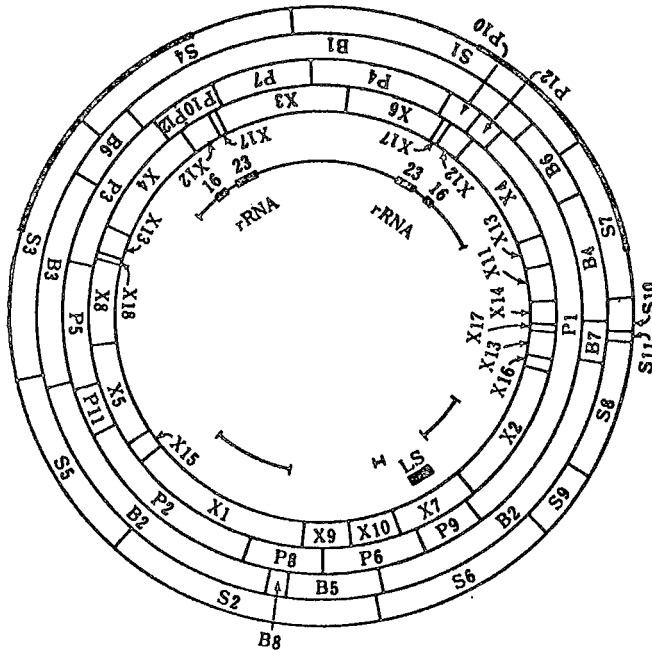


Fig. 1 Physical map of tobacco chloroplast DNA. Restriction fragments are S (SalI), B (BglII), P (PvuII) and X (XhoI). Thickened outerlines indicate inverted-repeat sequences. Thickened inner lines indicate the rRNA and the LS genes.

coding regions are highly conserved among higher plants and resemble quite well each other (about 90% homology). The positions in the gene corresponding to the 5' and the 3' ends of tobacco LS mRNA have been located by the S1 nuclease mapping procedure. The promoter sequence of the LS gene has homology with *Escherichia coli* promoter sequences; its terminator sequence is capable of forming a stem-and-loop structure. A sequence GGAGG, which is complementary to a sequence near the 3' end of tobacco chloroplast 16S rRNA and a putative ribosome binding site, occurs 6-10 bp upstream from the initiation codon. These observations suggest that molecular mechanisms of transcription and translation of chloroplast are similar to those of prokaryotes. In fact, we demonstrated the *E. coli* RNA polymerase can start the LS mRNA *in vitro* at the same position as that of *in vivo*.

We have also determined the nucleotide sequence of rRNA operon of tobacco chloroplast. Two sets of rRNA operon are located on inverted-repeat sequences (Fig. 1). The order of genes of rRNA operon is tRNA<sup>Val</sup>-16S rRNA-tRNA<sup>Ile</sup>-tRNA<sup>Ala</sup>-23S rRNA-4.5S rRNA-5S rRNA. The coding regions of 16S, 23S, 4.5S and 5S rRNA genes are 1485, 2804, 103 and 121 base pairs long. Nucleotide sequences of 16S and 23S rRNA have approximately 70% homology with those of *E. coli*. The 3' terminal region of 16S rRNA gene contains the sequence CCTCC which is complementary to sequence found at the 5' terminus of the LS mRNA. The 4.5S rRNA is found in the large subunit of the chloroplast ribosomes, and its nucleotide sequence has homology with that of 3' end of 23S rRNA. The nucleotide sequence of 5S RNA from tobacco chloroplast resembles well that from blue-green algae, which support the idea of the symbiotic origin of chloroplast genomes.

The region preceding the 16S rRNA gene contains tRNA<sup>Val</sup> (GAC) gene and promoter-type sequences similar to those which occur in *E. coli*. *E. coli* RNA polymerase can recognize these sequences and co-transcribes the tRNA<sup>Val</sup> and rRNA genes *in vitro*. The spacer region between the 16S and 23S rRNA genes is 2080 bp long and contain two tRNA genes; tRNA<sup>Ile</sup> and tRNA<sup>Ala</sup>. These two tRNA genes have large introns of about 700 bp in their anticodon loops. They are co-transcribed with 16S and 23S rRNA genes as a single precursor RNA of 8.2 kb. A long intron is also found in tRNA<sup>Val</sup> (UCA) gene which is located near the LS gene. The CCA sequence, which is coded for *E. coli* tRNA genes, is not found at 3' ends of tRNA genes of chloroplast. These observations reveal eukaryotic features of tRNA genes of chloroplasts.

## Evolution of *Triticum* and *Aegilops* viewed from the plasma type and chloroplast genome<sup>1)</sup>

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In order to characterize genetically the cytoplasms of all species of *Triticum* and *Aegilops*, two investigations have been conducted in 1982 with collaboration of Dr. Y. OGIHARA, Mr. H. TSUJIMOTO and many others. Some details of the results will be reported.

1. Investigation on the effects of the cytoplasms on various characters of 12 common wheats

Table. Classification of *Triticum* and *Aegilops* cytoplasms into different plasma types and chloroplast genome types

Plasma type	Cytoplasm (code number) of	Main characteristics expressed in common wheat		Chloroplast genome	Rubisco LS type
		Fertility spectrum	Other characteristics		
A	<i>boeoticum</i> (01), <i>monococcum</i> (16)	VIII	Reduced vigor, variegation, winter killing	1a	L
B	<i>longissima</i> (11), <i>dicoccoides</i> (21), <i>dicoccum</i> (22), common wheat (52)	I	Normal	7	H
C	<i>caudata</i> (02), <i>truncialis</i> (38)	VI	Partial pistillody, haploid induction	2	L
C <sup>u</sup>	<i>umbellulata</i> (03), <i>truncialis</i> (26), <i>biuncialis</i> (29, 37), <i>columnaris</i> (30), <i>triaristata</i> 4x(32) & 6x(54, 57)	IV	Reduced vigor, variegation, haploid induction	3	L
D	<i>squarrosa</i> (04), <i>cylindrica</i> (28)	I	Normal	9	L
D <sup>a</sup>	<i>ventricosa</i> (36)	I	Partial pistillody	1b	L
G	<i>crassa</i> 4x(35) & 6x(55), <i>juvenalis</i> (53), <i>vavilovii</i> (56)	I	Partial pistillody	1b	L
G	<i>aucheri</i> (09), <i>speltoides</i> (15), <i>nudiglumis</i> (23), <i>araraticum</i> (24), <i>timopheevi</i> (25), <i>zhukovskiyi</i> (51)	VII	Preharvest sprouting, reduced germination rate	5	H
M	<i>comosa</i> (05), <i>heldreichii</i> (06)	VIII	Reduced vigor, variegation	?	L
M <sup>o</sup>	<i>ovata</i> (31)	V	Delayed heading	6	L
M <sup>u</sup>	<i>uniaristata</i> (07)	II	Haploid induction	10	L
Mt	<i>mutica</i> (13)	II	Delayed heading, haploid induction	4	L
Mt <sup>a</sup>	<i>mutica</i> (14)	VIII	Haploid induction	4(?)	?
S	<i>speltoides</i> (08)	I	Almost normal	8	H
S <sup>b</sup>	<i>bicornis</i> (12)	I	Almost normal	1c	L
S <sup>l</sup>	<i>sharonensis</i> (10)	III	Reduced vigor	1d	L
S <sup>v</sup>	<i>kotschyi</i> (33), <i>variabilis</i> (34)	II	Haploid induction	1c	L

?: Not tested

Note) *Ae. searsii*'s chloroplast genome and Rubisco LS (Fraction I protein large subunit) are the same as those of S<sup>b</sup> and S<sup>v</sup> plasma types.

1) The work was supported by a Grant-in-Aid (No. 56440001) from Ministry of Education, Science and Culture, Japan.

About 500 nucleus cytoplasm (NC) hybrids, in which the nuclei of 12 common wheats are combined with the cytoplasm of 40 strains belonging to 32 species in all possible combinations, were grown together with normal lines of 12 common wheats in field under a split plot design, and their 16 characters were observed. Based on these results and some other supplementary data, the 40 cytoplasm could be classified into 16 plasma types, of which main features are given in the table above.

## 2. Electrophoretic patterns of restriction enzyme digests of chloroplast DNA

Chloroplasts (ct) were isolated from seedlings of self fertile NC hybrids having different cytoplasm. DNAs isolated from the chloroplasts were digested with four to seven restriction enzymes, and the so-called "restriction pattern" of the digests was analyzed by agarose gel electrophoresis. The results revealed that there are, at least, 13 chloroplast genomes distinctly differing from each other. The chloroplast genome of each cytoplasm is also given in the table.

Based on the results presented in the table and on other data, the phylogenetic relationships among the cytoplasm, and among the species of *Triticum* and *Aegilops* have been discussed.

### Effects of the nuclear-cytoplasmic interaction on grain protein in wheat

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In order to elucidate whether or not so called "cytoplasmic heterosis" (KIYHARA 1963) can be utilized for improvement of wheat protein (SASAKI *et al.* 1977, 1978), 4 alloplasmic ditelocentric (DT) line groups of Chinese Spring (CS) were analysed for grain protein and amino acid composition together with a number of plant characters. In this report the results obtained in 1979-1981 will be summarized on percentage grain protein (%P) and lysine content per protein (%L) of 95 DT and 5 euploid lines of CS each with one of the *T. aestivum* (original), *Ae. speltoides*, *Ae. ovata*, *Ae. variabilis*, *Ae. squarrosa* cytoplasm.

Alloplasmic CSDT lines were developed in the Laboratory by crossing between alloplasmic CS and respective CSDT lines, which were developed by E.R. SEARS (1954). A single-plant randomization design was used with three replications having 2 plants within each replicate block. Nitrogen was determined by the Carlo Erba Model 1300 Automatic Nitrogen Analyser. Total nitrogen  $\times 5.83$  was used to convert nitrogens to protein values. For each line 6 ground wheat samples were analysed 3 times within each sample. Amino acid composition was analysed by the Shimadzu HPLC Model LA3 Amino Acid Analyser using 3 samples for each line, with 2 determinations within each sample. Appropriate statistical adjustments were made to analyse the combined data of three different years.

Table 1. Relative genetical effects<sup>1)</sup> on percentage grain protein (%P) and lysine content per protein (%L) of ditelocentric lines (DT) of chinese spring (CS) with eu- or alloplasm

DT <sup>2)</sup> line	Cytoplasm										Mean	
	<i>T. aest.</i>		<i>Ae. spelt</i>		<i>Ae. ovata.</i>		<i>Ae. variab.</i>		<i>Ae. squar.</i>			
	%P	%L	%P	%L	%P	%L	%P	%L	%P	%L	%P	%L
1BS	— <sup>3)</sup>	—	3.3	-.49†	—	—	-0.8	-.18	2.5	-.10	1.6	-.26†
1DL	3.6	-.40†	4.3†	-.05	—	—	—	—	6.1†	.42†	4.6†	-.01
2AS	3.8	.02	5.8†	.14	2.8	-.15	3.3	-.03	5.1†	.55†	3.9†	.11
2BL	2.5	-.14	3.2	.44†	3.4	-.07	0.3	.14	—	—	1.7	.09
2DS	0.8	-.25	6.1†	-.11	—	—	1.8	.19	—	—	2.9	-.06
3AL	2.6	-.21	0.1	-.16	—	—	—	—	0.9	.60†	1.2	.08
3DL	—	—	-0.6	-.24	1.6	-.43†	0.8	-.19	2.2	-.28	1.1	-.28†
4DL	1.5	-.28	1.0	-.10	-0.2	-.06	-0.5	-.20	-0.1	-.20	0.4	-.17†
5AL	1.6	-.13	2.1	-.45†	2.6	-.05	1.7	-.20	7.5*	.45†	3.1	-.08
5BL	—	—	-0.2	-.10	3.0	-.08	4.4	.28	5.7†	.08	3.3	.05
5DL	3.2	-.19	-0.1	.01	—	—	2.2	-.23	2.4	.50†	1.9	.02
6AS	6.7*	.02	7.7*	-.33	—	—	2.4	-.02	3.8	-.14	5.1*	-.12
6DS	4.9†	-.31	5.0†	-.32	1.4	-.02	—	—	4.1	.26	3.9†	-.10
7BL	-0.3	0.6	-2.8	-.35	-1.7	-.16	0	.28	—	—	-1.2†	-.04
7DS	3.9	-.41†	6.3*	-.56†	1.8	-.23	1.2	-.01	—	—	3.3	-.30†
Euploid	0 <sup>4)</sup> (12.3)	0 <sup>4)</sup> (2.55)	1.4	-.09	0.8	-.05	1.3	.11	4.4†	.18	1.6 <sup>4)</sup>	.03 <sup>4)</sup>
Mean	2.4 <sup>4)</sup>	-.15 <sup>4)</sup>	2.2	-.14	1.0*	-.09	1.2*	.02*	3.3†	.17*	2.1	-.04

- 1) Differences in corrected %P and %L between the CS euploid in its own cytoplasm (%P 12.3, %L 2.55) and each line were used as relative genetical effects.
- 2) Only those DT lines including at least one line having significant genetical effect on one character are shown.
- 3) —: no data available.
- 4) As controls for each line, DT mean and cytoplasm mean, euplasmic euploid, euploid mean and cytoplasm mean were used, respectively.
- 5) \* and † : significant at the 5% level by the Tukey's test and the Duncan's Multiple Range test, respectively.

The %P means of CSDT lines examined varied from 11.1% for DT7BL with *speltooides* cytoplasm to 23.8% for DT2AS with *speltooides*, and most of them were higher than that of the CS euploid with *aestivum* cytoplasm. However, since the %P were correlated negatively with yield component characters, the %P were adjusted by the regression based on the seed fertility. A relative genetical effect of each line on %P was estimated by the difference between each line and the CS euploid in its own cytoplasm (Table 1). The genetical effect of 5.1% for DT6AS was significantly larger than that of 1.6% for the control CS euploid by the Tukey' test. This indicates that the long arm of CS chromosome 6A may carry the inhibiting gene or genes for protein content. That the cytoplasm means of *ovata* and *variabilis* were significantly lower than that of the control *aestivum* suggest that both cytoplasms may decrease %P of CSDT lines though that of the CS euploid with these cytoplasms was not low comparing with the euplasmic euploid. The interaction effects between CSDT lines and cytoplasms were also found in some lines such as

DT6AS with *aestivum*, 6AS and 7DS with *spelloides*, 5AL with *squarrosa* cytoplasm.

In a similar way the genetical effect of each line on %L was calculated (Table 1). As for %L only cytoplasmic effects were significant of *variabilis* and *squarrosa*, but neither nuclear or nuclear-cytoplasmic interaction effects.

However, by the Duncan's Multiple Rang test, which is not severe as the Tukey's test, genetical effects of some lines became significant in either or both %P and %L. These results indicate that several genes and interactions with smaller effects than those already mentioned may also be involved. Further studies in detail are necessary to clarify the above mentioned points including the role of *squarrosa* cytoplasm which seems to increase both %P and %L of certain genotypes of CS, in wheat protein improvement.

We are grateful to Dr. K. NISHIKAWA and Drs. H. FUKASAWA, K. TSUNEWAKI for providing us with the source seeds of CSDT lines and with the cytoplasm source line seeds, respectively. A part of this work was supported by a Grant-in-Aid from the Ministry of Education, Science and Culture, Japan and from the International Atomic Energy Agency, Vienna.

### Reproductive barriers in wild diploid wheats

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In diploid wheats two wild species, *Triticum boeoticum* Boiss. and *T. urartu* Tum., have been recognized. In order to clarify reproductive barriers involved in interspecific F<sub>1</sub> hybrids which might be occurred in several developmental stages, crossability, viable seed formation, germinability, hybrid weakness, and pollen and embryo sac fertility were observed. In this experiment 120 strains of *T. boeoticum* collected from Armenia, Turkey, Iraq, Iran, Lebanon, and Greece, and 16 strains of *T. urartu* from Armenia, Turkey, Iran, and Lebanon were used.

The intraspecific F<sub>1</sub> hybrid plants among five tester strains of *T. boeoticum* from Armenia, Turkey, Iraq, Iran, and Greece had high pollen fertility (more than 90%), that was estimated from the percentage of stainable good pollens, and high seed fertility (more than 80%), that was estimated from the percentage of seed set in the lowest floret of the spikelets. Similarly, F<sub>1</sub> plants among 16 strains of *T. urartu* from four different geographic areas had more than 60% pollen fertility and more than 40% seed fertility (Table 1). These results suggest that each diploid wheat throughout its distribution area was homogeneous with respect to genic factors affecting the fertility of F<sub>1</sub> plants, and that, therefore, no significant reproductive barriers existed within each species.

On the contrary, all interspecific crosses used *T. boeoticum* as a female parent gave small, plumped, but viable seeds. However, among 82 reciprocal crosses used *T. urartu* as a female parent, 66 cross combinations gave slender and shrivelled inviable seeds, and

Table 1. Pollen and seed fertility of F<sub>1</sub> plants among 16 strains of *T. wartsu* using 5 strains as testers

Area (Strain No.)	No. of strains	Pollen fertility (%)							Seed fertility (%)						
		100	90	80	70	60	50	40	100	90	80	70	60	50	40
Armenia (199-1)	16	8	5	2	1				1	5	2	4			4
Turkey (199-5)	15	10		1	4					4	2	2	3		4
(199-13)	15	13	2						2	5	6	2			
Lebandon (199-7)	15	11	4						2	6	5	1	1		
Iran (199-9)	14	7	5	2					7	1	1	4	1		

Table 2. Pollen and seed fertility of F<sub>1</sub> hybrids derived from *T. boeoticum* × *T. wartsu* (Lebanon)

Area (No. of strains)	Pollen fertility (%)											Seed fertility (%)										
	100	90	80	70	60	50	40	30	20	10	0	100	90	80	70	60	50	40	30	20	10	0
Armenia (11)								1		10											1	10
Turkey (30)		2					3	6	4	15				1	1	3	5	20				
Iraq (18)							1	2	2	13								3	15			
Iran (5)										5												5
Lebanon (6)										6												6
Greece (2)										2												2
Total		2					4	9	6	51				1	1	3	9	58				

Table 3. Embryo sac fertility of F<sub>1</sub> hybrids backcrossed by *T. boeoticum* or *T. wartsu*

Cross combination	Seed set (%)	Germination rate (%)
<i>(boeoticum</i> × <i>wartsu</i> ) × <i>boeoticum</i> or <i>wartsu</i>		
(3620 × 199-1) × 3620 or 199-1	25	74
(3620 × 199-5) × 3620 or 199-5	22	82
(3620 × 199-8) × 3620 or 199-8	33	78
(8120 × 199-5) × 8120 or 199-5	22	86
(8396 × 199-5) × 8396 or 199-5	27	83
(8396 × 199-16) × 8396 or 199-16	25	84
<i>(wartsu</i> × <i>boeoticum</i> ) × <i>boeoticum</i> or <i>wartsu</i>		
(199-1 × 3620) × 3620 or 199-1	28	89
(199-5 × 3620) × 3620 or 199-5	25	50
(199-8 × 3620) × 3620 or 199-8	33	83
(199-5 × 8120) × 8120 or 199-5	35	88
(199-5 × 8396) × 8396 or 199-5	30	81
(199-16 × 8396) × 8396 or 199-16	28	70

only 16 cross combinations gave some shrivelled viable seeds. Crossability between two species did not differ significantly in the reciprocal crosses using *T. boeoticum* or *T. wartsu* as a female parent, but germinability differed significantly.

In these interspecific crosses the F<sub>1</sub> hybrids derived from certain cross combinations showed hybrid weakness, indicating yellow coloration at the tip of leaves that begins at

about tenth day after germination and poor adventitious root development. At 25°C in the greenhouse, those plants stopped their growth at three-leaf stage and died soon. However, in the experiment field most of them survived, and a clear distinction between them and normal plants could not be recognized during the winter season. But at tillering stage necrosis started at the tip of leaves and progressed to the whole plant finally. All of them came to die before the maturing stage. The reciprocal crosses also gave the same symptoms. This results suggests that hybrid necrosis is not caused by cytoplasmic factors, but by two complementary genes designated as *Ned*<sub>1</sub> and *Ned*<sub>2</sub>. Of 120 strains of *T. boeoticum* examined, 48 had *Ned*<sub>1</sub> gene and all 16 strains of *T. urartu* had *Ned*<sub>2</sub> gene.

The pollen and seed fertility in the F<sub>1</sub> hybrids between the remaining 72 strains of *T. boeoticum* and *T. urartu* from Lebanon are shown in Table 2. The table indicates that pollen and seed fertility are generally very low, with the exceptions of hybrids involved one Armenian, eleven Turkish, and three Iraqi strains. The pollen fertility was 20–30%, 20–80%, and 20–40%, respectively, and the seed fertility was all 10–50%. The embryo sac fertility of F<sub>1</sub> hybrids used *T. boeoticum* and *T. urartu* as female parents is shown in Table 3. Embryo sac fertility was estimated from both the percentage of the seed set in pollinated florets and the germination rate of obtained seeds. In all cases the seed set was about 30% and the seed germination rate was about 80%. All data indicate that the reproductive barriers, i.e., inviable seed formation, hybrid weakness, reduced pollen and embryo sac fertility, play as significant isolation mechanisms between two wild diploid wheat species.

### Cytogenetical studies of B-chromosomes in a weedy rye, *Secale afghanicum*

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*Secale afghanicum* (Vavilov) Roschev. is a weedy species closely related to cultivated rye, *Secale cereale*. B-chromosomes (Bs) found in the original samples of *S. afghanicum* were investigated cytogenetically (AKITA & SAKAMOTO 1982). In this brief article two particular characteristics, such as the pairing frequencies of Bs and the effects of Bs on the pairing of A-chromosomes (As) at MI of pollen mother cells (PMCs), are reported. The results were also compared with the cytogenetic characteristics of Bs found in *S. cereale*.

The original samples of *S. afghanicum* used in this study were collected in Afghanistan in 1978. Of eight populations from five original samples, six included the plants with 1B–4Bs. The standard type of Bs had subterminal centromere. Though two other karyotypes of Bs, probably derived from standard ones, were also found, only the plants with the standards Bs were examined.



The data on the B-chromosome pairing at MI of PMCs is shown in Table 1. In the plants with two Bs, two univalents of Bs were observed in more than 60% of PMCs examined. Most bivalents of Bs were rod ones, whereas ring bivalents were found to be much rare (less than 5% of bivalents). The pairing frequency of Bs in *S. afghanicum* was not different from that in several reports on Bs of *S. cereale* (MÜNTZING 1945, SARVELLA 1959). In the plants with 4Bs or 6Bs, the pairing behavior of Bs was characterized by much rare formation of multivalents at MI of PMCs. Such multivalents were usually formed as a Y-shaped trivalent (in two 4B plants) or an X-shaped quadrivalent (in a 6B plant).

To examine the effects of Bs on the pairing of As, total 71 plants in five strains derived from the same plant, were observed at MI of PMCs, as shown in Table 2. The plants without Bs usually showed seven bivalents. On the other hand, the plants with 2Bs in the

Table 1. Mean B-chromosome pairing at MI of PMCs

Culture number	No. of Bs	No. of plants examined	I	II		III	IV
				rod	ring		
8045	2Bs	15	1.31	0.33	0.02	—	—
8046	2Bs	10	1.20	0.38	0.01	—	—
	4Bs	1	1.78	0.89	0.06	0.11	—
8049	2Bs	3	1.25	0.34	0.03	—	—
	4Bs	2	1.93	0.83	0.04	0.11	—
	6Bs	1	1.61	1.84	0.10	0.14	0.03
1001*	2Bs	9	1.40	0.30	0.00	—	—
	4Bs	1	2.86	0.54	—	0.02	—
1004**	2Bs	5	1.63	0.18	—	—	—

\* Artificially intercrossed progeny between plants of 8045.

\*\* Artificially intercrossed progeny between plants of 8046.

Table 2. Mean A-chromosome pairing at MI of PMCs

Culture number	No. of Bs	No. of plants examined	No. of cells observed	I	II		No. of Xta
					rod	ring	
8045	0B	7	350	—	0.73	6.27	N.O.
	2Bs	15	750	0.15	1.40	5.52	N.O.
8046	0B	4	200	0.02	0.92	6.07	N.O.
	1B	2	100	0.14	1.38	5.55	N.O.
8049	2Bs	10	500	0.14	1.32	5.61	N.O.
	2Bs	3	150	0.05	1.58	5.39	N.O.
	4Bs	2	100	0.26	1.81	5.06	N.O.
	6Bs	1	80	0.18	1.86	5.05	N.O.
1001	0B	7	350	0.09	1.01	5.94	13.15
	1B	1	100	0.08	1.65	5.31	12.73
	2Bs	9	450	0.30	2.06	4.79	12.24
	4Bs	1	100	0.50	2.72	4.03	11.64
1004	0B	3	150	0.08	1.89	5.07	12.61
	1B	1	50	0.16	2.02	4.90	12.30
	2Bs	5	250	0.42	2.31	4.48	12.02

N.O.; Not observed.

same strain showed the increase of univalents, the decrease of ring bivalents and the increase of rod bivalents. Moreover, the number of paired arms per cell and the mean chiasma frequency per cell of As in the 2B plants decreased and became variable. For instance, in culture no. 1001 the mean chiasma frequency of each 0B plant ranged from 12.88 to 14.08 (13.51 on the average). On the other hand, that of each 2B plant ranged from 10.84 to 13.26 (12.24 on the average). In general, the number of ring bivalents and the mean chiasma frequency of As in the 2B plant were lower than those without Bs. Similar results were reported by TSUMOTO & SASAKI (1972), while, contradictory results were also found by ZEČEVIC & PAUNOVIC (1969) in *S. cereale*.

**An analysis of meiotic chromosome pairing by a mathematical model  
in *Aegilops speltoides* and its hybrids carrying  
B-chromosomes**

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An analysis was made on *Ae. speltoides* ( $2n=14$ ; SS) and its hybrids involving the diploid wheat ( $2n=14$ ; AA) or the tetraploid wheat of the emmer group ( $2n=28$ ; AABB), some of which included various numbers of B-chromosomes. The mathematical model used in the present study assumes that the process of chromosome pairing consists of two successive, independent events. The first event is the association of chromosomes as entire units, and the second event is the chiasma formation between arms of the associated chromosomes, occurring with probability  $a$  and  $c$ , respectively (DRISCOLL *et al.*, 1979). The probability with which one randomly sampled chromosome is involved in various configurations was calculated and is indicated in Table 1. In the calculation about the *Ae. speltoides*  $\times$  diploid wheat hybrids, it was assumed that there was an interchange per cell which might show a chain configuration. Varying numbers of cells were observed for each material, and one chromosome was randomly sampled from each pair of homologues in the case of *Ae. speltoides* and from each cell in the cases of its hybrids involving either the diploid or the tetraploid wheat. Estimation of the two parameters,  $a$  and  $c$ , was performed by the method of maximum likelihood, based on the polynomial distribution (Table 1).

The observed and expected frequencies of various configurations are shown in Tables 2 and 3. These results indicate that the present model of chromosome pairing explains the observations reasonably well. Therefore it is worthwhile to compare the values of the estimates of the two parameters ( $\hat{a}$  and  $\hat{c}$ ) among these materials. The values of  $\hat{a}$  and  $\hat{c}$  of the OB-class of *Ae. speltoides* were 1.0 and 0.93, respectively. The presence of various numbers of B's (1 to 5) had almost no effects on these values (Table 2). When OB-classes were considered, there were no significant differences concerning the value of  $\hat{a}$  between

homologous chromosome pairing in *Ae. speltoides* and homoeologous chromosome pairing in the hybrids (Tables 2 and 3), but the values of  $\hat{c}$  of the latter (0.49 and 0.70) were smaller than the former (0.93). Therefore reduced amount of pairing generally seen between homoeologues may result from the failure of chiasma formation. When B's were present in the hybrids, further reduction in the amount of homoeologous chromosome pairing was

Table 1. The probability of sampling one chromosome randomly and the estimates of  $a$  and  $c$

Material	Configuration & Probability	Estimates of $a$ and $c^*$
<i>Ae. speltoides</i>	Univ.: $(1-a)+a(1-c)^2$ Rod biv.: $2ac(1-c)$ Ring biv.: $ac^2$	$\hat{a} = \frac{x+y}{x+y+z} \cdot \frac{1}{\hat{c}(2-\hat{c})}$ $\hat{c} = \frac{2x}{2x+y}$
<i>Ae. speltoides</i> × diploid wheat	Univ.: $(1-a)+a(1-c)(7-6c)/7$ Rod biv.: $ac(1-c)(13-c)/7$ Ring biv.: $5ac^2/7$ Triv.: $3ac^3(1-c)/7$ Quadriv.: $2ac^3/7$	$\hat{a} = \frac{v+w+x+y}{v+w+x+y+z} \cdot \frac{1}{\hat{c}(13/7-6c/7)}$ $6(v+w+y)\hat{c}^3$ $-(110v+104w+13x+26y)\hat{c}^2$ $+(442v+351w+182x+104y)$ $\hat{c}-169(2v+w+x) = 0$
<i>Ae. speltoides</i> × tetraploid wheat	Univ.: $(1-a)+a(3-2c)^2/9$ Rod biv.: $4ac(1-c)/3$ Ring biv.: $2ac^2/9$ Triv.: $2ac^3/3$	$\hat{a} = \frac{w+x+y}{w+x+y+z} \cdot \frac{1}{\hat{c}(4/3-4c/9)}$ $\hat{c} = \frac{3w+3x}{3w+3x+2y}$

\*  $v, w, x, y,$  and  $z$  are the observed frequencies of quadrivalent, trivalent, ring bivalent, rod bivalent, and univalent, respectively.

Table 2. Observed and expected chromosome pairing per cell in *Ae. speltoides* and the estimates of  $a$  and  $c$

B-class	No. of chromosomes	O/E*	I	II		$\hat{a}$	$\hat{c}$
				Rod	Ring		
0	2100	O	0.06	0.94	6.0	1.0	0.93
		E	0.07	0.91	6.1		
1	1610	O	0.01	0.75	6.2	1.0	0.94
		E	0.05	0.79	6.2		
2	1050	O	0.05	0.81	6.2	1.0	0.94
		E	0.05	0.79	6.2		
3	350	O	0.08	1.2	5.7	1.0	0.90
		E	0.14	1.3	5.7		
4	280	O	0.30	1.5	5.3	0.99	0.87
		E	0.37	1.6	5.3		
5	210	O	—	0.63	6.4	1.0	0.95
		E	0.04	0.67	6.3		

\* Observed or expected.

Table 3. Observed and expected chromosome pairing per cell in the hybrids and the estimates of  $\hat{a}$  and  $\hat{c}$

Cross	B-class	No. of chromosomes	O/E	I	II		III	IV	$\hat{a}$	$\hat{c}$
					Rod	Ring				
<i>Ae. speltoides</i> × diploid wheat	0	250	O	4.0	3.2	1.1	0.30	0.14	1.0	0.49
			E	4.1	3.1	1.2	0.24	0.12		
	1	300	O	12.4	0.72	0.09	—	—	0.39	0.18
			E	12.3	0.74	0.06	0.02	0.00		
<i>Ae. speltoides</i> × tetraploid wheat	0	565	O	6.5	2.7	1.2	2.1	(0.14)	0.95	0.70
			E	6.7	2.8	1.1	2.2			
	1	522	O	13.9	2.9	0.22	0.25	(0.03)	1.0	0.24
			E	14.8	2.6	0.13	0.27			
	2	172	O	18.7	0.98	0.18	—	—	0.41	0.22
			E	18.7	0.98	0.05	0.09			

observed. In these cases, the values of  $\hat{a}$  also decreased in addition to further decrease of the  $\hat{c}$  values (Tables 2 and 3). Therefore it is concluded that presumably both association and chiasma formation of homoeologous chromosomes are suppressed by the presence of B's and that those of homologous chromosomes are not affected by their presence.

### Identification of the genomes involved in reciprocal translocations in the wild tetraploid wheats

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Several reciprocal translocations of chromosomes have been found among strains of the two wild tetraploid wheats, *Triticum dicoccoides* Körn. and *T. araraticum* Jakubz. (KAWAHARA & TANAKA 1977, 1978, 1981, TANAKA *et al.* 1979). Of these reciprocal translocation chromosome types, E<sub>1</sub> of *T. dicoccoides* and T<sub>1</sub> of *T. araraticum* were assumed to be the original chromosome structures of the two species, respectively (KAWAHARA & TANAKA 1981). The other types differ from the original ones by one or two translocations. In order to compare the degree of structural differentiation of chromosomes belonging to the different genomes, chromosomes involved in these translocations were identified through crossing experiments with wild diploid wheats, *T. boeoticum* Boiss. Materials used were nine strains of *T. dicoccoides* of types E<sub>1</sub>, E<sub>2</sub>, E<sub>3</sub>, E<sub>4</sub> and E<sub>6</sub>, one of *T. timopheevi* (T<sub>1</sub>) and nine of *T. araraticum* (T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub>, T<sub>4</sub>, T<sub>5</sub> and T<sub>6</sub>). They were crossed by diploid wheats and the chromosome pairings in F<sub>1</sub> hybrids were observed.

Of nine hybrids of *T. dicoccoides* with diploid wheats (AAB hybrids), one hybrids

involving  $E_6$  type formed a quadrivalent per cell. Since no quadrivalent was formed in hybrids of  $E_1$  type, this indicates that the translocation between  $E_1$  and  $E_6$  type are located on two different chromosomes belonging to the A genome. Similarly, identification of genomes involved in other translocations was made by comparing the occurrence of multivalents in PMCs of hybrids with original  $E_1$  types with that of hybrids with the other types. Two translocations between  $E_1$  and  $E_2$  or  $E_3$  were considered to be those between chromosomes of the B genome. The translocation between  $E_1$  and  $E_4$  possibly involve chromosomes of the A genome and that of the B genome. In hybrids of *T. timopheevi* or *T. araraticum* with diploid wheats (AAG hybrids), more multivalents were formed in PMCs than in AAB hybrids. Because chiasma frequencies of AAG hybrids were similar to those of AAB hybrids, this would indicate that the A genome of *T. timopheevi* and *T. araraticum* is structurally differentiated from that of diploid wheats. One or two translocations were found between original  $T_1$  and the other types. But there was little difference in the occurrence of multivalents between hybrids of  $T_1$  type and those of the other types. Probably, these translocations involve no chromosome belonging to the A genome. Based on the present observation and the occurrence of multivalents among translocation types, chromosomes involved in several translocations in *T. dicoccoides* or *T. araraticum* were identified.

The present results show that the chromosomes of the B or G genomes are more frequently involved in translocations in the tetraploid wheats than those of the A genome. Of five translocations in *T. dicoccoides*, three were between chromosomes of the B genome, one was between those of the A genome and one was between the A and B genomes. In *T. araraticum*, all the eight translocations identified were between chromosomes of the G genome. It is concluded that both of the B and G genomes of the tetraploid wheats show higher degree of variability in chromosome structures than that of the A genome.

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### Classification of tetraploid wheats based on the response to *Aegilops squarrosa* cytoplasm

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A total 100 strains of tetraploid wheats were investigated on the genetic compatibility of their genomes to the cytoplasm of *Aegilops squarrosa*. These tetraploid wheats were

classified into three types of responses to *squarrosa* cytoplasm, as AB, AG and AB' type (Table 1).

Table 1. Different response of tetraploid wheats to *Aegilops squarrosa* cytoplasm appeared in seed morphology and seedling development of F<sub>1</sub> in the crosses to (*squarrosa*) AABB+1D lines.

Response type of male parent	Seed morphology		Seedling development	
	Normal	Abortive	Normal plant	Midget plant
AB type	(sq.) AABB+1D	(sq.) AABB (zygotic lethal)	(sq.) AABB+1D (2n=29)	—
AG type	(sq.) AABG+1D (sq.) AABG	—	(sq.) AABG+1D (sq.) AABG (2n=29 & 28)	—
AB' type	(sq.) AABB'+1D (sq.) AABB'	—	(sq.) AABB'+1D (2n=29)	(sq.) AABB' (2n=28)

Genetic analysis proved that two nuclear factors affected by the compatibility to *squarrosa* cytoplasm. One of them was a factor for plant vigour, which might affect the development or ability of mitochondrion. The other was considered as a factor for the development of plastid. The incompatibility of the latter factor to *squarrosa* cytoplasm was manifested as follows; gametic sterility in male with lack of starch, zygotic lethality caused by degeneration of endosperm, and chlorophyll variegation in plant due to abnormal development of chloroplast.

Majority of Emmer wheats (AABB genomes), *Triticum turgidum* (3 strains), *T. durum* (11 strains), *T. polonicum* (2 strains), *T. orientale* (2 strains), *T. aethiopicum* (3 strains), *T. isphahanicum*, *T. dicoccum* (17 strains) and *T. dicoccoides* (4 strains from Palestine and 5 strains from Zagros Mts. region) showed the zygotic lethality under the cytoplasmic background of *Ae. squarrosa*. When these strains were crossed to the lines having the genetic constitution of (*squarrosa*)AABB+1D, many abortive seeds (zygotic lethal) were produced and their genetic constitution was presumed to be (*squarrosa*)AABB. On the other hand, all F<sub>1</sub> seedlings from non-abortive (normal) seeds developed to normal plants in growth with the chromosome number of 2n=29 indicating the genetic constitution of (*squarrosa*)AABB+1D, although these plants were partially male sterile due to the gametic sterility of (*squarrosa*)AB. Genomes of these species were incompatible to *squarrosa* cytoplasm, and classified as AB type on the basis of their response to *squarrosa* cytoplasm.

Timopheevi wheats (AAGG genomes, 7 strains of *T. timopheevi*, 6 strains of *T. araraticum* collected in Trans-Caucasia and 18 strains of *T. araraticum* collected in Zagros Mts. region) were completely compatible to *squarrosa* cytoplasm. When the (*squarrosa*)AABB+1D lines were pollinated with these Timopheevi wheats, no abortive seeds were observed, and all the F<sub>1</sub> seedlings developed to normal plants in growth irrespective of their chromosome number such as 2n=28 or 2n=29. These strains were classified as AG type.

Three endemic species of cultivated Emmer wheat (9 strains of *T. persicum*, 4 strains of

*T. pyramidale* and 4 strains of *T. palaeocolchicum*) and 4 strains of wild tetraploid wheat collected in Zagros Mts. region (3 strains classified as *T. dicoccoides* and 1 strain classified as *T. araraticum*) showed incomplete compatibility to *squarrosa* cytoplasm. When these strains were used as male parents in the cross experiments, no abortive seed were produced on the (*squarrosa*)AABB+1*D* plants. These strains were differed from other Emmer wheats (AB type) in the response to *squarrosa* cytoplasm, because they did not exhibited the zygotic lethality in the nucleo-cytoplasmic combinations with *squarrosa* cytoplasm. However, they were also different from AG type, because they showed segregation in the development of F<sub>1</sub> seedlings. The F<sub>1</sub> seedlings with 1*D* chromosome (2n=29) grew to normal plants, but the F<sub>1</sub> seedlings without 1*D* chromosome (2n=28) turned to be midget plants with extreme reduction of plant vigour and severe chlorophyll variegation under low temperature. These strains were classified as AB' type.

No difference in the response type were observed among the strains belonging to the same species, with exception of four strains of wild tetraploid wheat collected in Zagros Mts. region. These exceptional strains were classified as AB' type in this experiment, regardless they were named as *T. dicoccoides* or *T. araraticum*. However, in respect of cytological and morphological classification, TANAKA & KAWAHARA (1976) suggested that two of these strains (KU-8821A and KU-8821C) were intermediate strains between AB genome species and AG genome species.

The present results indicate that the genetic differentiation in compatibility to cytoplasm among tetraploid wheats correspond to the phylogenetic differentiation of tetraploid wheats. Such a correspondence suggests that the compatible relation between nucleus and cytoplasm, as revealed in the present experiment, may have some relation with the differentiation of species in wheat and the relatives.

The author wish to express his sincere gratitude to Dr. M. TANAKA of Kyoto University and Dr. G. Kimber of University of Missouri for supplying the seeds of tetraploid wheat strains used in the present experiment.

**Genetic variations in the AABB genome extracted from *Triticum durum-Elytrigia elongata* (*Agropyron elongatum*, 2n=14) chromosome addition lines**

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A zygotic lethal mutation was found in the chromosome complements of A or B genome within a *durum-Elytrigia* chromosome addition line (Ono, Nakamura and Kido, 1981). The alien chromosome addition plants carrying the zygotic lethal gene(s) set seeds normally, but majority of them (ca. 80%) were shrivelled and ungerminated. The remaining plump seeds

germinated well and grew to maturity. The matured plants had an alien chromosome with no exception and set shrivelled seeds with frequencies similar to their parent plants. This indicates that when the alien chromosome is incorporated in the *durum* wheat carrying the lethal gene(s), the lethal effect is compensated by the added chromosome. This type of change, if occurred, makes the alien chromosome indispensable for the addition line and seems likely to demonstrate an instance of chromosomal differentiation which must have occurred in the course of evolution of polyploid species.

Mochizuki (1962) who produced the *durum-Elytrigia* addition lines reported that the added *Elytrigia* chromosomes more or less associated or paired with their corresponding wheat chromosomes at metaphase I, except for  $e_4$  chromosome. Under such condition, gene transfer to wheat from *Elytrigia* may have occurred by means of recombination between added chromosome and chromosome of the recipient.

To examine if any differences in phenotypical expressions have occurred between the *durum* wheat and the extracted AABB genome plants from each addition line for the last 20 years since they were produced, the extracted AABB plants were compared with normal *durum* wheat in several morphological characters, mainly quantitative ones, such as plant height, tiller number, ear length, ear density, seed fertility and so on. The extracted AABB plants were obtained from self-pollinated progenies of monosomic addition plants which were produced by crosses between monosomic *durum* and *Elytrigia* chromosome

Table 1. Frequencies of AABB plants extracted from eight sources monosomic additions.

Monosomic line	No. of plants examined	No. of chromosomes		
		28	29	30
e1	105	89	16	0
e2	104	94	10	0
e3	108	104	4	0
e4t <sup>s</sup> -1	91	84	7	0
e4t <sup>s</sup> -2	110	98	12	0
e5	72	28	34	10
e6	100	94	6	0
e7	98	91	7	0

Table 2. Chromosome configurations at metaphase I of the monosomic addition.

Monosomic line	No. of PMC observed	Chromosome configurations (%)					
		14 <sup>v</sup> +1 <sup>v</sup>	13 <sup>v</sup> +3 <sup>v</sup>	1 <sup>iv</sup> +13 <sup>v</sup>	1 <sup>iv</sup> +12 <sup>v</sup> +2 <sup>v</sup>	1 <sup>iv</sup> +12 <sup>v</sup> +1 <sup>v</sup>	1 <sup>v</sup> +12 <sup>v</sup>
e1	200	94.0	5.5	0.5			
e2	150	91.3	8.7				
e3	150	92.0	3.3		0.7	4.0	
e4t <sup>s</sup>	150	86.0	14.0				
e5	200	40.5	1.5	54.5	1.5		2.0
e6	50	94.0	4.0			2.0	
e7	50	90.0	4.0			6.0	



Table 3. Ear length (cm) of *durum* wheat and the AABB plants extracted from the monosomic additions.

AABB plants from	Block number				Mean
	I	II	III	IV	
<i>durum</i> wheat	9.13	9.10	9.10	9.58	9.23
e1	10.14	9.25	9.26	9.47	9.53
e2	10.08	9.56	9.44	9.79	9.72
e3	9.87	9.47	8.83	8.92	9.27
e4t <sup>s</sup> -1	10.56	10.83	9.33	9.58	10.08
e4t <sup>s</sup> -2	9.56	9.33	8.79	10.09	9.44
e6	10.39	11.66	9.84	10.19	10.52
e7	10.08	9.89	9.50	9.64	9.78

Analysis of variance				
Item	DF	SS	MS	F
Line	7	5.2814	0.7544	4.2028**
Block	3	2.4418	0.8139	4.5342**
Error	21	3.7708	0.1795	
Total	31	11.4940		

disomic addition, except for e<sub>6</sub>, in which the monosomic addition plants were obtained directly from disomic addition, because of their chromosomal instability.

Table 1 records the frequencies of AABB plants derived from eight sources of monosomic additions and Table 2 shows the chromosome configurations at metaphase I of those monosomic additions. In an experimental field, every 80 plants of the normal *durum* and the seven extracted AABB plant, except for ones obtained from e<sub>6</sub>, were divided into four blocks, comprising eight randomized plots assigned to each of the eight kinds of AABB plants to be compared. The extracted AABB from e<sub>6</sub> addition and *durum* wheat were grown in a greenhouse.

Statistical analysis of the observation in ear length was presented in Table 3 as an example, revealing a significant difference at 1% level among AABB plants under test.

### III. Editorial Remarks

#### Announcement for Future Issues

WIS No. 57 will be planned for publication in October, 1983, Manuscripts for this issue are most welcome and accepted any time, not later than August 31, 1983.

WIS is open to all contributions regarding methods, materials and stocks, ideas and research results related to genetics, breeding and cytology of *Triticum*, *Aegilops*, *Secale*, *Haynaldia* and related genera. Manuscripts should be typewritten (double-space) in English, and submitted with duplicates. One article should not exceed five printed pages, including two textfigures (smaller than 7×7 cm<sup>2</sup>). Lists of stocks are exempted from this page limit. Authors receive 50 reprints of their contributions free of charge. Extra copies are printed by order at cost price. Communications regarding editorial matters should be addressed to:

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*The Managing Editor*

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### *Explanation of the Figure on the Cover*

The symbol mark of the 6th International Wheat Genetics Symposium. The Symposium is planned to be held at Kyoto, Japan in Nov. 28 - Dec. 3, 1983. See WIS No. 55 for the details.

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